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Some Chemical Aspects of Resistance to Cercospora Leaf Spot in Sugar Beets[1]

MERLE HARRISON, MERLE G. PAYNE AND JOHN O. GASKILL[2]

Received for publication November 16, 1960

Cercospora leaf spot, potentially a highly destructive disease of the sugar beet (*Beta vulgaris* L.) in many parts of the world, is being partially controlled by the use of resistant varieties. Leaf-spot resistant commercial varieties currently available in the United States, under conditions of moderate disease exposure, provide relatively satisfactory control; however, in seasons or localities in which conditions are especially favorable for development of the causal organism (*Cercospora beticola* Sacc.), such varieties may be seriously damaged by the disease. Consequently, varieties with improved resistance are urgently needed. The investigations reported in this article where undertaken in an attempt to explore the mechanism of leaf-spot resistance and to point the way toward improved selection techniques.

Review of Literature

In order for an infection to be successfully completed there must be an interaction among the host, the pathogen, and the environment, which favors the pathogen and its parasitic establishment. It is only in the last few decades that a pathogen's preference for certain varieties of the same species has been investigated. In the majority of cases investigated, the factors responsible for disease resistance have fallen into one of the following categories: disease escape, mechanical exclusion, lack of an essential substance for development of the pathogen and presence or production of a substance toxic to the pathogen (6, 7, 21, 22).[3]

A possible mechanism of resistance of young leaves to Cercospora leaf spot was postulated regarding stomata size when it was observed that the pathogen had to gain its entrance through the open stomata (16), but there is adequate evidence that even seedlings with small stomates can be infected (20). In another investigation of resistance to Cercospora leaf spot no significant differences were found in regard to the size and number of stomates on resistant and susceptible varieties (8).

[1] Cooperative investigations of the Colorado Agricultural Experiment Station and the Crops Research Division, Agricultural Research Service of the United States Department of Agriculture. Approved by the Colorado Agricultural Experiment Station for publication as Scientific series Article No. S.S. 693.

[2] Instructor of chemistry and research assistant, associate professor of chemistry, Colorado State University, and plant pathologist, United States Department of Agriculture, respectively.

[3] Numbers in parentheses refer to literature cited.

Kovacs (13) found that the low incidence of local lesions on the leaves of a resistant sugar beet variety was correlated with the presence of diffusable inhibitors on healthy leaves. Fewer spores of *C. beticola* germinated on the resistant leaves and a correspondingly smaller percentage of the spores germinated in dew or water washings collected from resistant leaves. No attempt was made to identify or characterize the inhibitor.

From growth studies of *C. beticola* in pure culture it seemed unlikely that the resistant leaves could be lacking in any essential nutrient for the growth of the organism (4).

As early as 1911 Cook et al. (3) pointed out that at the point of penetration of a pathogen the action of polyphenoloxidases might bring about reactions which could inhibit the progress of the pathogen. Since that time there has been a great deal of literature published regarding the role of phenolic compounds in relation to disease resistance. Thoroughly investigated examples are those of resistance to onion smudge (14, 15) and resistance of potatoes to common scab (9, 10). In the first instance the resistance was found to be due to concentrations of catechol and protocatechuic acid and in the latter to chlorogenic acid. In the case of potato scab resistance, certain polyphenolic compounds were found to promote the development of a thicker suberized layer (17).

The polyphenolase enzyme may function in the disease resistance mechanism and has been found capable of being activated (1, 11, 12). In sugar beet leaves most of the enzymes are in the stable adsorbed condition on the plastids and autolysis of the plastids causes the enzymes to go into solution (18, 19).

Material

The eight inbred lines of sugar beets used in this investigation are listed in Table 1. The leaf-spot readings, furnished by the United States Department of Agriculture[4], were made in field plots in 1957 under severe, artificially induced leaf-spot exposure. On this scale 0.0 represents complete immunity while 10.0 represents complete defoliation.

The causal organism, *C. beticola,* was isolated from infected leaves, and single-spore cultures were used for toxicity studies. Potato-dextrose agar was used as the growth medium for the toxicity tests.

Experimental Design

The eight inbred lines were planted in the United States Department of Agriculture's Sugar Beet Greenhouse in Fort

[4] Acknowledgment is made to J. A. Elder, Agricultural Aid, United States Department of Agriculture, for conducting field tests and making leaf-spot readings.

Table 1.—Contributors of the inbreds and the relative leaf-spot readings.

Contributor	Contributor's number	Leaf-spot reading	Experimental line number
United States	US 201	0.5	1
Department of	52-437	7.5	7
Agriculture	52-335	1.0	3
The Great Western	GWI-29-55L	1.0	2
Sugar Company	GWI-20-55L	7.0	8
	GWI-21	8.0	6
American Crystal Sugar Company	ACS-I_1	1.3	4
Holly Sugar Corporation	5126-0	9.0	5

Collins, Colorado, September 22, 1959. Each line occurred in four plots. Each plot consisted of two eight-inch pots—each containing two plants—making a total of four plants of a single line per plot. A randomized-block experimental design was employed.

Methods

The four youngest fully expanded leaves on each plant were harvested January 15, 1960. The leaves, with petioles removed, were frozen immediately and stored in polyethylene bags in a freezer until analysis.

It was found that if the leaves were allowed to thaw at room temperature, the phenolic compounds were partially lost—apparently through oxidation by the polyphenolase enzyme. Hence, for extraction of phenolic compounds, the frozen leaves were placed in 95% ethanol and ground immediately in a Waring blender for three minutes. The concentration of the ethanol was kept above 80% to insure deactivation of the enzyme as soon as the cell was ruptured.

For the extraction, 15 grams of frozen leaves were ground in 75 ml of 95% ethanol and filtered through a Buchner funnel using vacuum. The residue was washed with an additional 25 ml of 95% ethanol. The ethanol solution was allowed to stand for one hour at room temperature and then refiltered through Whatman No. 50 filter paper. This solution was taken to dryness under partial vacuum with gentle heat and the residue taken up in 10 ml of distilled water. The aqueous solution was made to pH 2.5 using concentrated hydrochloric acid. The acidic solution was extracted three times with 10 ml portions of ether to remove some undesirable components, and the resulting solution was used for further experiments. For convenience this crude solution will be referred to as a phenolic extract.

The phenolic extracts and the oxidized phenolic extracts were tested for toxicity by placing a drop next to a 2 cm mat of *C. beticola* and observing the growth three days later.

The polyphenolase enzyme was isolated from the frozen leaves by allowing them to thaw and pressing to extract the juice. This juice was centrifuged, dialyzed at 4° C with a phosphate buffer at pH 6.7, lyophilized and separated electrophoretically in a phosphate buffer at pH 6.7 with 25 ml of glycerol added per liter. The polyphenolase fraction migrated toward the (–) electrode.

Analyses and Results

The causal organism grew well on many simple media, including potato-dextrose agar, Czapek's agar etc.; however, the best growth and conidia formation were obtained using a beet-leaf agar from either the resistant or susceptible lines. Conidia production was slight on media such as Czapek's agar, but could be increased many times by the addition of a small amount of riboflavin.

When the phenolic extract from a resistant line was applied to the organism growing in culture, there was very little inhibition, but when the phenolic extract was oxidized prior to application there was severe inhibition. The substance responsible for the inhibition was heat stable, allowing the solutions to be heated to remove the oxidizing agents and also to increase the concentration.

Figure 1 shows the effect of adding two drops of phenolic extract from the most resistant line which had been heated in a water-bath until it was concentrated five times (about 2 to 3

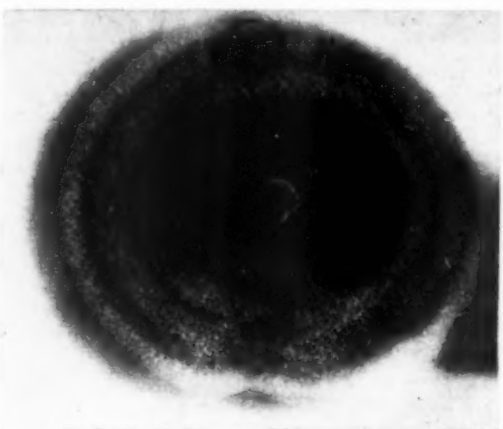

Figure 1.—The effect of adding two drops of concentrated phenolic extract to a culture of C. *Beticola*.

hours). The picture was taken three days after application and shows very little inhibition.

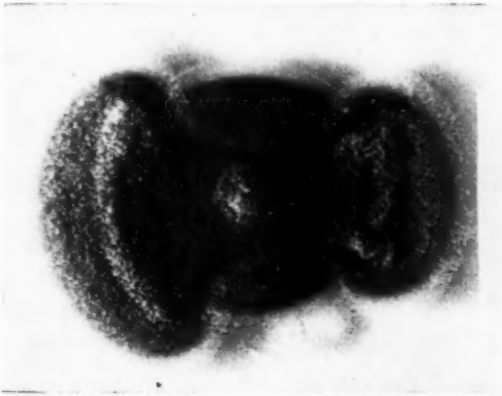

Figure 2.—The effect of adding, to a culture of *C. beticola*, two drops of concentrated phenolic extract which has been ozidized with hydrogen peroxide.

Figure 2 shows the effect of phenolic extract treated in a similar manner except that one drop of 30% hydrogen peroxide was added to 10 ml prior to heating. The water-bath treatment was sufficient to remove any excess hydrogen peroxide as indicated by testing with catalase enzyme. There was severe inhibition and even some existing growth was destroyed. This inhibition cannot be attributed to the interaction of the hydrogen peroxide and the hydrochloric acid because, when a water blank was treated with hydrochloric acid, hydrogen peroxide and heated, no inhibition was observed. When the extract from a susceptible line was oxidized and tested in a similar manner negligible inhibition was observed.

A similar inhibition to that in Figure 2 was observed when nitric acid was used instead of hydrochloric to bring the extract to pH 2.5, indicating that the inhibition must have been due to oxidation and not merely to hydrogen ion concentration. Other chemical oxidizing agents had similar effects and the extract could also be oxidized to a toxic substance using the polyphenolase enzyme.

Due to the low concentration of the phenolic extract, paper chromatography seemed to offer the best means of separation and characterization. Figure 3 shows a diagramatic composite of two dimensional chromatograms. The first dimension was run in a butanolacetic acid-water mixture[5], and the second in five

[5] 120 ml butanol, 30 ml acetic acid and 60 ml water.

percent acetic acid. Spot E represents ninhydrin positive compounds (mostly amino acids), spot D represents a sugar spot (mostly glucose), while A, B and C represent phenolic spots. When spots A, B and C were isolated from a number of chromatograms, oxidized with hydrogen peroxide, concentrated and tested for toxicity on the organism; spot A was the only one

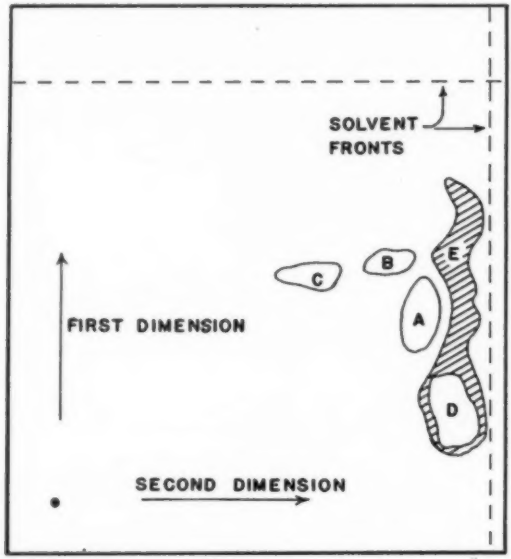

Figure 3.—A diagramatic composite of two dimensional chromatograms.

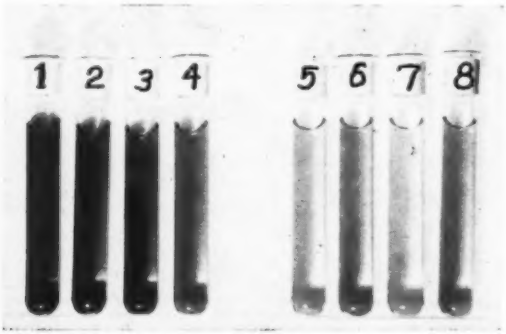

Figure 4.—The relative intensities of the violet color obtained by treating representative samples of sugar beet leaf extracts from lines 1 to 8 with Arnow's reagent.

exhibiting toxicity. It was found that Arnow's reagent (2) modified for paper chromatography[a] reacted selectively with spot A to produce an immediate reddish-brown color in the presence of ammonia vapors.

Since the phenolic compound A in Figure 3 was the only substance in the extract that reacted appreciably with Arnow's reagent, the relative concentration of the compound responsible

Table 2.—Optical density measurements for 32 greenhouse plots of sugar beets.

Line number	Leaf-spot reading (field)	Plot number (greenhouse)	Optical density	Average of 4 plots
1	0.5	8	2.30	1.480
		11	1.60	
		20	0.80	
		29	1.22	
2	1.0	1	1.60	0.885
		15	0.40	
		19	0.71	
		28	0.83	
3	1.0	7	1.60	0.925
		14	1.50	
		21	0.50	
		30	0.10	
4	1.3	4	0.77	0.530
		10	0.50	
		22	0.48	
		32	0.37	
5	9.0	6	0.22	0.2775
		9	0.61	
		17	0.15	
		27	0.13	
6	8.0	5	0.50	0.475
		16	0.38	
		18	0.44	
		26	0.31	
7	7.5	2	0.24	0.200
		12	0.15	
		23	0.20	
		31	0.21	
8	7.0	3	0.38	0.225
		13	0.22	
		24	0.11	
		25	0.19	

[a] 1 part water, 2 parts 95% ethanol, and 1 part Arnow's reagent (10 g sodium nitrite and 10 g sodium molybdate in 100 ml water).

for the potential inhibition of the pathogen could be determined in solution. For this analysis 0.5 ml of the phenolic extract, 1.0 ml Arnow's reagent (2), and 25 ml water were added to an Evelyn tube. One ml of 10% sodium hydroxide was added and the resulting violet color read at 515 mμ on a Beckman Model B spectrophotometer. Figure 4 shows a representative sample from each of the eight lines. The violet color is much more intense in the resistant than in the susceptible lines.

The results of this method of analysis are given in Table 2, and Table 3 gives the analysis of variance for those data. From these tables it can be seen that: 1) there is a significant difference at the 1% level between resistant and susceptible lines in the amount of phenolic compound present, 2) the resistant lines contain varying concentrations of the phenolic compound, 3) line number 1 contained the highest concentration of the phenolic compound, and 4) there are significant differences at the 1% level among replications.

Table 3.—Analysis of variance for optical density measurements presented in Table 2.

Source of variation	Mean square	Degrees of freedom
Resistant vs. susceptible lines	3.6700**	1
Within resistant lines	0.6161**	3
Within susceptible lines	0.0342	3
Replications	0.5085**	3
Lines vs. replications	0.1152	21

**Significant at the 1% level.

In an attempt to characterize compound A from Figure 3, identical chromatograms were run and dyed with various reagents. These results are presented in Table 4.

Table 4.—Color reactions of compound A in Figure 3.

Reagent	Color reaction
Folin-Denis	blue
Sulfanilic acid	red
P-nitraniline	purple
Arnow's	reddish-brown
Ferric chloride	green-blue which fades rapidly
Ultra-violet light	very faint purple

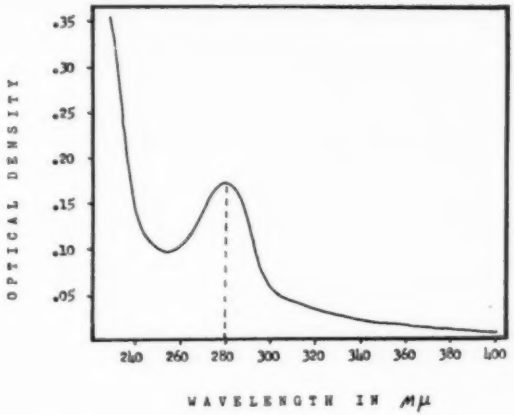

Figure 5.—The ultraviolet absorption spectrum of the potentially toxic phenolic compound.

Figure 5 gives the ultraviolet absorption curve obtained by means of a DU Beckman spectrophotometer using substance isolated from spot A Figure 3. For this isolation many chromatograms were run, one chromatogram was dyed, and spot A isolated from the other chromatograms. These spots were eluted with water to give a solution for the analysis. There is a maximum absorption at 280 mμ indicating a phenolic substance which has no conjugation with the benzoid structure.

The phenolic substance responsible for the inhibition of *C. beticola* is water-soluble and heat-stable. When the extract was extracted with ether, ethyl acetate and amyl alcohol and oxidized there was no inhibition to the organism in the ether or ethyl acetate layers, only slight inhibition in the amyl alcohol layer, with most of the toxicity remaining in the aqueous portion.

No differences could be found in the electrophorograms from resistant and susceptible leaves. The portion containing the polyphenolase activity moved farther toward the cathode than any other zone of protein. This portion accounted for a considerable percentage of the entire cytoplasmic protein in the leaves. The activities of the enzymes obtained from resistant and susceptible lines were very nearly the same, although activation could have taken place during extraction. Because of the relative abundance and activity of the polyphenolase enzyme in the susceptible as well as the resistant lines, it appeared unlikely that the enzyme could be the limiting factor in any reaction controlling resistance.

In another experiment washings were taken from line number 1 and a susceptible line (not included in the greenhouse series) by washing the leaves with a washbottle filled with distilled water and collected in a polyethylene bag. The washings were taken from 50 leaves (both upper and lower surfaces) and were about 150 ml each. These were concentrated to 10 ml in a water bath and examined chromatographically. In the washings from the resistant line there was a faint spot corresponding to spot A in Figure 3. It gave the same color reactions as spot A and was entirely lacking in the washings from the susceptible line.

Discussion

From the various color reactions with reagents on chromatograms and from the ultraviolet absorption data the compound responsible for the toxicity to *C. beticola* appears to be a single ortho-dihydroxy phenolic compound which has no conjugation in the side groups. This compound cannot be a simple dihydroxy compound as evidenced by its partition with water and ether. The simple phenolic compounds are soluble in ether. However, since the potentially toxic compound is water soluble it must not contain any long aliphatic side-chains.

A possible mechanism of disease resistance is as follows: The organism entering the cell (or for that matter any general injury) might cause the polyphenolase enzyme to act on the phenolic compound producing toxic substances. Since there is evidence that this enzyme can be activated by various agents: that in the sugar beet the enzymes are adsorbed on the plastids: and that autolysis of the plastids causes the enzymes to go into solution, injury could release, activate or merely mix the enzymes with the cell contents. The oxidized phenolic compound then would inhibit the pathogen directly aand perhaps exhibit a secondary inhibitory effect by stimulating the walling-off of the wound. Cunnngham (5) reported that the zone between the dead cells of the leaf-spot and the healthy cells is characterized by mature cells which have become meristematic and solidly packed. The fungus hyphae are never present in this zone or beyond. Furthermore, if the phenolic compound is presentt on the surfaces of the leaves of resistant lines it may inhibit the germination of the conidia of the pathogen.

Although it is risky to isolate an artifact from plant material and reason that it was the same and underwent the same reactions in the living cell, the results of this investigation strongly indicate that selection of individual plants for leaf-spot resistance can be performed effectively by chemical means. The most

promising technique appears to be that involving optical density determinations for solution samples prepared with Arnow's reagent.

Summary

A phenolic compound was isolated from leaves of sugar beets which, when oxidized, is extremely toxic to *C. beticola* growing in pure culture. This compound was found to be more concentrated in the leaves from resistant inbred lines than in the leaves from susceptible inbred lines.

This compound appears, from chromatographic, solubility, and ultra-violet absorption data, to be an ortho-dihydroxy phenol with one or more side groups. These side groups probably contain no conjugation with the benzoid structure.

The polyphenolase enzyme isolated from the leaves does not appear to be the controlling factor in producing resistance.

The compound responsible for the toxicity was found in the washings from leaves of a resistant line growing in field tests, but was lacking in washings obtained from a susceptible line.

A possible mechanism of leaf-spot resistance in sugar beets is described.

The results indicate the feasibility of selecting individual sugar beet plants for leaf-spot resistance on the basis of chemical determinations on leaf samples.

Literature Cited

(1) Allen, T. H., Ray, O. M. and Bodine, J. H. 1938. Enzymes in orthopteran ontogensis. VI. Autocatalytic nature of *in vivo* formation of protyrosinase. Soc. for Expt. Biol. and Med. Proc. 39: 549-552.

(2) Arnow, L. E. 1937. Colorimetric determination of the components of 3,4-dihydroxyphenylalanine-tyrosine mixtures. J. Biol. Chem. 118: 531-537.

(3) Cook, M. T., Bassett, H. P., Thompson, F. and Taubenhaus, J. J. 1911. Protective enzymes. Science 33: 624-629.

(4) Coons, G. H. and Larmer, G. F. 1929. The physiology and variations of *Cercospora beticola* in pure culture. Mich. Acad. of Sci., Arts, and Letters 11: 75-104.

(5) Cunningham, H. S. 1928. A study of the histological changes induced in leaves by certain leaf spotting fungi. Phytopathology 18: 717-751.

(6) Gaumann, E. A. 1950. Principles of plant infection. p. 245-439. Hafner Publishing Company, New York.

(7) Horsfall, J. G. and Dimond, A. E. 1959. Plant pathology, an advanced treatise. Vol. I. p. 435-467, Academic Press, New York.

(8) HOSOKOWA, S. 1958. Annual report of sugar beet experiments in 1958. p. 46-60. Hokkaido National Agricultural Experiment Station, Division of Field Crops, Japan.

(9) JOHNSON, G. and SCHALL, L. A. 1952. Relation of chlorogenic acid to scab resistance in potatoes. Science 115: 627-629.

(10) JOHNSON, G. and SCHAAL, L. A. 1957. Chlorogenic acid and other orthodihydricphenols in scab-resistant russet Burbank and scab susceptible triumph potato tubers of different maturities. Phytopathology 47: 253-255.

(11) KENTEN, R. H. 1957. Latent phenolase in extracts of broad-bean (*Vicia faba* L.) leaves. 1. Activation by acid and alkali. Biochem. J. 67: 300-307.

(12) KENTON, R. H. 1958. Latent phenolase in extracts of broad-bean (*Vicia faba* L.) leaves. 2. Activation by anionic wetting agents. Biochem. J. 68: 244-251.

(13) KOVACS, A. 1955. Uber die Ursachen der unterschiedlichen Resistenz der Zuckerrubensorten gegen *Cercospera beticola* Socc. Phytopathol. Zeitschrift 24: 283-298.

(14) LINK, K. P., ANGELL, H. R. and WALKER, J. C. 1929. The isolation of protocatechuic acid from pigmented onion scales and its significance in relation to disease resistance in onions. J. of Biol. Chem. 81: 369-375.

(15) LINK, K. P. and WALKER, J. C. 1933. The isolation of catechol from pigmented onion scales and its significance in relation to disease resistance in onions. J. of Biol. Chem. 100: 379-383.

(16) POOL, V. W. and MCKAY, M. B. 1916. Relation of stomatal movement to infection by *Cercospora beticola*. U. S. Department of Agriculture, J. Agr. Research 5: 1011-1038.

(17) SIMONDS, A. O., JOHNSON, G. and SCHAAL, L. A. 1953. Comparative effects of catechol, some related compounds, and other chemicals on suberization of cut potato tubers. Botan. Gaz. 115: 190-195.

(18) SISAKYAN, N. M. and KOBYAKOVA, A. M. 1948. Activity and condition of enzymes in plastids. Biokhimiya 13: 88-94.

(19) SISAKYAN, N. M. and KUVAEVA, E. B. 1949. Polyphenoloxidase and peroxidase activity of isolated plastids. Doklady Akad. Nauk. S.S.S.R. 62, 121-124 (1948) in Chemical Abstracts v. 43 (1), col. 1083.

(20) VESTAL, E. F. 1933. Pathogenicity, host response and control of Cercospora leaf-spot of sugar beets. Iowa Agr. Expt. Sta. Research Bull. 168: 45-70.

(21) WALKER, J. C. 1957. Plant pathology. p. 584-585. McGraw-Hill Book Co., Inc., New York.

(22) WALKER, J. C. and STAHMANN, M. A. 1955. The chemical nature of disease resistance in plants. Annual Rev. of Plant Physiol. 6: 351-366.

Planter Tests and Seed Preparation Using Monogerm Sugar Beet Seed[1]

R. D. BARMINGTON[2]

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Introduction

The goal of sugar beet mechanization is complete elimination of hand labor. For all practical purposes this goal has been reached in harvesting the crop but hand labor in the spring is still necessary as an aid to mechanization and is sometimes the only satisfactory way of doing the thinning and weeding job. Elimination of hand labor could be accomplished by three factors, namely, better seedling stands, more accurate distribution, and adequate weed control.

An important factor contributing to a satisfactory seedling stand is the planting equipment. If truly precision seed metering and row distribution can be combined with a consistently high percentage of seedling emergence in the field, then the plant population problem will be solved. At the same time improved mechanical and chemical weed control techniques must be developed.

Precision planting of monogerm sugar beet seed on its present state of development is very difficult due to some unfavorable physical characteristics. Accurate metering and distribution of the seed in a soil environment favorable for germination is largely an engineering problem and is one with which this paper deals.

The search for new ideas in planting mechanisms continues and some interesting ideas show up from time to time. It is the opinion of the author that some seed modification is the most promising solution when it is used in combination with the best possible planting mechanisms. The planter mechanisms used in 1959 field trials will be discussed first and the seed modification later.

Planting Equipment and Experimental Method Used

For the 1959 field trials a rear mounted tool bar of sufficient length to accommodate six planter units was used. Each planter mounted on the tool bar had some features that were different from each other unit. All the units, except one, were equipped to plant either bare monogerm seed or coated monogerm seed. A total of 10 farmers' fields were used in the tests. In each field one round was made using bare monogerm seed and one using coated monogerm seed.

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[1] Scientific Journal Series No. 648, Colorado Agricultural Experiment Station.

[2] Associate Agricultural Engineer, Agricultural Engineering Section, Colorado Agricultural Experiment Station, Colorado State University, Fort Collins, Colorado.

Figure 1.—The experimental machine used in farmers' fields consisted of six different planting units. Left to right they are: Stanhay, Silver, John Deere No. 70, experimental International, commercial International and Milton.

All the planting was done in five working days over a seven-day period. Storms delayed general planting in the Fort Collins area but the experiments were planted as timely as possible and at random, the selection depending upon which fields were ready. Actual planting dates were from April 23 to 29 inclusive.

Mounted on the tool bar from left to right the planting units were: Stanhay, Silver, John Deere, experimental IHC, commercial IHC, and commercial Milton (Figure 1). The Stanhay is an English planter which has a belt metering device in which the seed cells are cut all the way through the belt. The seed enters one side of the cell and is discharged from the opposite side. It has a rotating seed cut-off and a shoe opener. Seed cells for bare seed were .165 inch in diameter and for coated seed the cell diameter was .250 inch.

The Silver unit was an experimental model with an unusual seed metering device. It used a vacuum to pick up the seed. The seed wheel, with short nozzles extending radially from the circumference, passed through seed in the hopper as a vacuum was applied to the nozzles. Vacuum held the seed until it reached the point of discharge where positive pressure was applied. On this particular unit a small gasoline engine was used to drive a vacuum pump. The furrow was formed by the combination of a single disc and a shoe opener. Although a number of imperfections were observed on this model the idea is challenging and deserves further study and development. On this model the nozzles were

not large enough to hold coated seed so only bare monogerm seed was planted in the test.

The John Deere Number 70 unit was one of the latest designs supplied by the manufacturer. After the unit was received, tapered steel washers were added to the disc hub bosses to change the pinch point of the discs to a lower position, thus eliminating most of the small ridge of soil normally left at the bottom of the furrow by disc openers. Cover knives were used to pull soil over the planted row behind the disc openers and in front of the presswheel. The seed plate for bare seed was .105 inch thick and cell diameter was .168 inch and for coated seed they were .150 by .187 inch in thickness and diameter.

The experimental International planter had the same hopper and plates as the commercial machine while the furrow opener and related parts were different. The disc casting was constructed so that the seed tube came down in front of the disc hub instead of behind it. A small spring steel rod extended from the front of the disc casting down between the discs to the bottom of the furrow to remove the small ridge of soil in the bottom of the furrow. A small seed pressure wheel ran between the back of the disc openers directly on the seed to firm the seed in the soil before the seed was covered. A single cover knife was used to pull soil over the seed before the presswheel ran over it. Two designs of zero pressure presswheels were used and were changed at each end of the field to give complete data on the effect of the two designs. Seed plates for bare seed were .125 inch thick with .156 inch diameter cells and for coated seed they were .178 inch thick with a cell diameter of .218 inch.

The commercial International planter used the same size seed plates as the experimental machine and was equipped with cover knives which are available as optional equipment. No special equipment or attachments were used with this machine.

The Milton planter was standard in every respect. Seed cells in this planter are not round holes and are given in 64ths of an inch in three dimensions. The first dimension is along the circumference of the seed wheel, the second figure is parallel to the axis of rotation and the third dimension is perpendicular to the axis. Cell size for bare seed was 10-8½-8½ and for coated seed it was 12-11-11.

Another unit which was not available when this field test was planted, but was included in a late season test, was an experimental belt planter built by the Great Western Sugar Company (Figure 2). The seed cells in this machine are holes

drilled part-way through a rubber belt. The belt passes through the seed body on a slope so that a seed cut-off is not necessary in the hopper. The seeds enter and are discharged from the same side of the belt. Minimum seed damage results with this system and the seed is discharged near the bottom of the furrow which seems to be an advantage. Some functional difficulties were experienced but the principle shows some merit and should be given further consideration.

Seed Used for the 1959 Field Test

The bare seed used in the 1959 field tests was screened through a 10/64 inch round hole sieve and over an 8/64 inch sieve giving it a seed ball count of 54,800 seed pieces per pound. After coating to form the pellets, the size was increased to the range of 10/64 to 12/64 inch and the weight increased 5.2 times the original weight.

Pelleted seed is easy to meter in a planter and gives uniform seed distribution in the furrow when used with properly sized plates. However, the cost of manufacturing, transporting, and storing greatly increases the final seed cost.

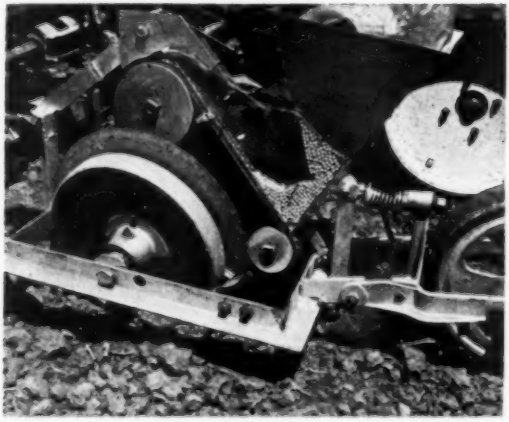

Figure 2.—Experimental Great Western belt planter using pelleted seed.

Field Observations

As a whole, all the units operated with a minimum of trouble in the field. As could be expected the experimental units required more careful attention and more adjustment than com-

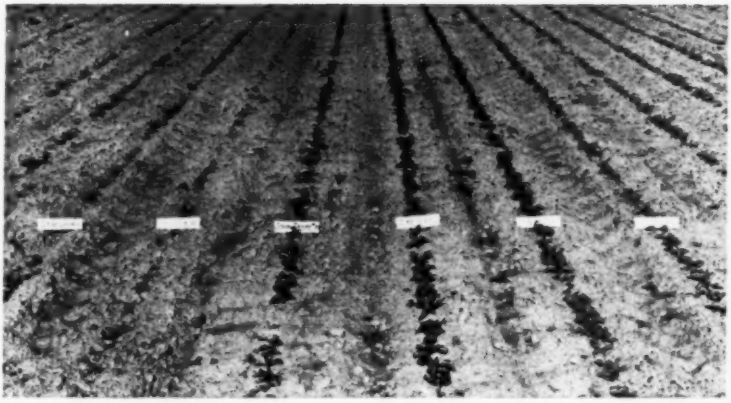

Figure 3.—Earlier germination and more vigorous plants resulted from the planters having cover knives to push soil over the seed in front of the presswheel. This difference disappeared as the season advanced and the fields became drier.

mercial units. After the plots in five fields had been planted it was necessary to dismantle and clean the vacuum pump which had become clogged with beet seed dust and oil. If this type of planter should become commercial, it would be necessary to incorporate an air cleaner between the planting units and the source of vacuum.

In one field the soil was wet enough that the furrow openers turned up wet, sticky soil which built up on the presswheels of all the planters. Occasionally this situation becomes a problem where zero pressure rubber presswheels are used. Scrapers can not be used effectively on this type of tire and when mud builds up on the wheel it does not function properly. Build-up of mud is also a problem with the small seed pressure wheel running on the seed between the disc furrow openers under certain conditions. However, this was not serious in any of the fields in the 1959 tests but did occur in a test conducted in 1950.

In the first four fields planted, the beet seedlings emerged more uniformly and showed more early vigor from the planters using cover knives to pull soil over the seed ahead of the presswheels (Figure 3). After considerable adjustment for planting depth and presswheel alignment on the experimental Silver machine, two of the fields showed more seedling vigor and earlier germination of seedlings from this machine compared to other planters in the test. Earlier work has shown that in the

Rocky Mountain area a definite relationship exists between soil firmness around the seed, soil moisture in the seed zone, and seedling emergence (1)[3]. These conditions are affected by the type of presswheel used. Improved germination can often be achieved by merely increasing the pressure on the presswheel. However, no clear cut over-all advantage was evident for any of the machines from the standpoint of early emergence and vigor when all of the fields are considered together.

Field Data and Method of Collection

Planter performance in the field is measured by the percent of seeds which germinate and the distribution of plants in the row. Rapid germination of seeds and early vigor of plants are desirable factors and are associated with planter design, but are usually recorded as an observation rather than numerical values.

In the first field planted there were twelve treatments and thirteen in all the rest. Only one type of presswheel was used on the experimental International planter in the first field.

The collection of field data was started as soon as the seedlings were large enough to count. The location for each count was selected at random. A measuring device 100 inch-spaces long resembling a long slender ladder with cross bars every inch was laid over the row. Two men working together recorded the number of plants in each one-inch space. A total of ten 100-inch counts was recorded for each treatment in each field. The following averages were calculated: 1) inches containing beets per 100 inches, 2) percent emergence, 3) percent of inch spaces containing singles, 4) doubles, and 5) multiple plants.

Laboratory Work Connected With the Field Test

With the exception of the experimental Silver machine, all planter drives and plates were selected in the laboratory to give a seed spacing of approximately 1½ inches. The Silver machine was set by the manufacturer to give approximately the same spacing. Percent cell fill was calibrated in the laboratory before going to the field in attempt to get cell fill as near 100 percent as possibe with the seed plates available. Seed ball distribution was determined in the laboratory by catching the seed on a greased board. A planter which placed all the seeds in the theoretically correct places was given a score of 100 and a score of zero if only half of the seeds were correctly placed (2). In the field the weight of seed planted by each machine and the length of rows were recorded. Percentage cell fill is obtained by comparing the theoretical number of seeds planted with the actual number.

[3] Numbers in parentheses refer to literature cited.

Summary and Conclusion

The average results obtained from each machine and a comparison between bare monogerm seed and coated monogerm seed are summarized in Table 1. Differences from farm to farm reflected change in moisture and seedbed preparation. Soils varied from very sandy to very heavy and moisture conditions from very wet at the beginning to very cloddy and dry on the surface by the end of the week. Most of the beets germinated from rains which came after planting except for the beets planted during the first two days. Due to wet spring weather most fields were worked when too wet, resulting in cloddy and loose seedbeds. Since all the planters were used on all the farms, differences between planters in the over-all results must be due to the effect of the planter itself. The purpose in using as many farms and field conditions as possible was to increase confidence in the final results. It is felt that considerable confidence can be placed in the data and this experiment will be useful for measuring progress in the development of planting equipment for monogerm seed.

It is possible to plant monogerm seed with present planting equipment. However, to realize the maximum potential from monogerm seed some method must be found to do a more accurate job of seed distribution. This may be done in one of three ways: 1) coating the seed to form a smooth, round and dense pellet, 2) polishing the seed to remove excess rough, corky material, thereby making the seed more nearly spherical and higher in density, and 3) new, improved, or refined planter design.

In this test coated seed gave an average of 88.57 percent singles while bare seed produced 83.47 percent. In this range, it is the author's opinion that the improvement in percent of single plants resulting from coated seed is not worth the added cost of coating. Results of a mid-summer planting using bare untreated seed, bare polished seed, and polished and coated seed showed that bare polished seed emerged three days sooner than untreated seed. The polished and coated seed emerged next with unpolished bare seed last. This would indicate that there may be some side benefits from planting bare polished seed. As a result of this observation, a more comprehensive study of seed polishing and seed sizing is under way with a two-fold purpose in mind. First, polished seed screened to closer limits should be much easier to meter in the planter, resulting in more uniform seed distribution. Second, if polishing the seed will hasten germination without detrimental effects to the seedlings, more vigorous and uniform stands will result in the field.

Table 1.—Summary of 1959 field results from planters using bare and coated monogerm sugar beet seed. Results for each planter are averages of ten farmers' fields.

	8-10/64" Bare Monogerm Seed						10-12/64" Coated Monogerm Seed					
Planter	**Beet inches per 100"**	**% Emergence**	**% Cell fill**	**% Single**	**% Doubles**	**% Multiples**	**Beet inches per 100"**	**% Emergence**	**% Cell fill**	**% Single**	**% Doubles**	**% Multiples**
Stanhay	24.90	53.22	79.3	86.47	12.57	0.95	25.97	51.72	76.2	92.08	7.47	0.44
Silver	27.88	49.76	108.6	87.04	11.42	1.53						
John Deere No. 70	30.43	58.04	95.7	89.52	9.63	0.86	30.67	54.83	94.5	91.26	8.40	0.84
Exp. IHC-Chev. p.w.	37.10	57.58	129.4	78.86	18.68	2.46	30.55	53.84	103.6	85.63	13.77	1.62
Exp. IHC-Rib p.w.	35.15	54.56	127.6	79.69	18.03	2.28	30.24	52.51	103.6	87.09	11.75	1.17
Commercial IHC	36.63	58.97	135.4	77.80	19.37	2.84	30.51	54.91	102.0	85.79	12.69	1.47
Milton	29.78	50.85	113.1	84.88	13.55	1.67	27.34	48.58	106.3	89.57	9.90	0.53
Total	221.98	382.98	788.4	584.26	103.25	12.59	175.28	316.39	586.2	531.42	63.98	6.07
Average	31.70	54.71	112.62	83.47	14.75	1.80	29.21	52.73	97.7	88.57	10.66	1.01

LSD .05 Level		LSD .01 Level	
Percent Emergence	5.828	Percent Emergence	4.928
Percent Cell Fill	8.478	Percent Cell Fill	6.872
Percent Singles	2.453	Percent Singles	2.074
Percent Doubles	2.117	Percent Doubles	1.790

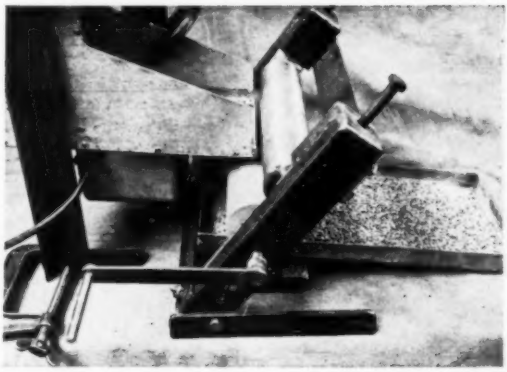

Figure 4. Large sizes of monogerm seed were decorticated fairly successfully by passing the seed between a steel roller and a rubber roller. Seed was fed to the rollers by a vibrating feed to space the seeds on the rollers.

Seed Polishing Possibilities

The seed used in this test was all monogerm which had been polished in a laboratory machine consisting of a series of emery wheels on an inclined shaft with baffles to cause the seed to mix as it went through the machine. This system was not completely satisfactory since experimental evidence shows that the seeds lie flat against the emery wheels and only a small part of the corky material is removed.

Later attempts at polishing monogerm seed have been made by both the Great Western Sugar Company and Colorado State University. Monogerm seed of larger sizes was decorticated quite successfully at Colorado State University by passing the seed between a steel roller and a rubber roller (Figure 4). In this system the steel roller supports the seed and the rubber roller pushes the corky corners down breaking them off. However, this device does not remove the cork from small seed.

Another idea which has been tried is the ball mill principle. To test the idea, a gem polisher (Figure 5) was used with one-quarter inch steel nuts, glass marbles, wood blocks, and screened gravel as grinding elements (Figure 6). The quality of polishing was very good, but the batch process was too slow, at least on a small scale. A tomato seed polisher was also tried and the results were fairly good but it has the same disadvantages of the gem polisher.

Table 2.—Cell fill and seed damage for various seed and cell sizes.

J. D. No. 70 beet planter and seed data. Seed spaced 1½ inches apart in the row.

Planting speed	Size of seed	Plate cell diameter	Cell depth	Percent cell fill	% Cracked through 6/64" screen	Observations
2½ mph	6-7/64	.125	.080	102.46	6.91	
2½ mph	7-8/64	.154	.080	104.06	6.21	Good performance
2½ mph	7-8/64	.154	.080	104.76	6.81	Good performance
2½ mph	8-9/64	.159	.128	117.06	7.21	
2½ mph	8-9/64	.173	.103	111.64	8.16	Slight diff. in the plate made a
2½ mph	8-9/64	.168	.105	98.66	5.49	noticeable diff. in perform.
2½ mph	9-10/64	.177	.127	98.45	3.75	
2½ mph	9-10/64	.177	.127	104.33	4.47	
3 mph	6-7/64	.125	.080	103.53	8.27	
3 mph	7-8/64	.154	.080	101.00	3.65	Best performance
3 mph	8-9/64	.159	.128	113.83	5.30	
3 mph	8-9/64	.159	.101	83.36	5.66	
3 mph	8-9/64	.173	.103	103.66	8.69	
3 mph	8-9/64	.168	.080	64.83	10.26	
3 mph	8-9/64	.168	.105	97.80	6.80	
3 mph	9-10/64	.177	.127	97.33	4.72	

Table 2.—Continued

IHC beet planter and seed data. Seed spaced 1½ inches apart in the row.

Planting speed	Size of seed	Plate cell diameter	Cell depth	Percent cell fill	% Cracked through 6/64" screen	Observations
2½ mph	6-7/64	.128	.078	98.10	7.40	
2½ mph	6-7/64	.156	.081	145.45	No Data	
2½ mph	6-7/64	.147	.097	166.25	No Data	
2½ mph	7-8/64	.156	.081	98.39	3.90	Best performance
2½ mph	7-8/64	.147	.097	118.50	No Data	
2½ mph	7-8/64	.140	.082	96.30	3.70	
2½ mph	8-9/64	.156	.081	58.66	2.95	This size seed and plate combination gave the most difficulty.
2½ mph	8-9/64	.173	.122	136.90	No Data	
2½ mph	8-9/64	.147	.097	63.76	No Data	
2½ mph	8-9/64	.157	.125	113.24	8.86	
2½ mph	9-10/64	.173	.122	94.20	8.40	
3 mph	6-7/64	.128	.078	97.00	5.70	
3 mph	7-8/64	.140	.082	94.53	5.35	Best performance
3 mph	8-9/64	.157	.125	112.50	6.18	Most difficulty

The Great Western Sugar Company is also working on some other ideas. Prospects for successful seed polishing in the near future appear promising and will contribute materially to successful planting of bare monogerm seed.

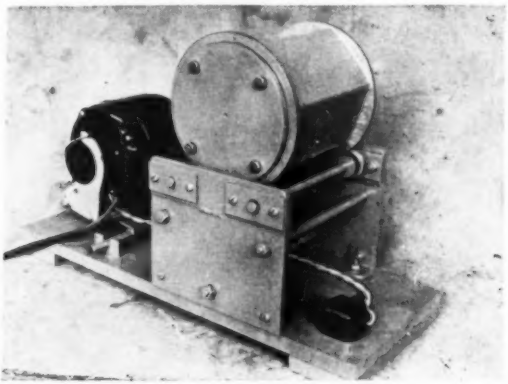

Figure 5. A gem polisher was used to test the ball mill principle of polishing monogerm sugar beet seed.

Figure 6. Grinding elements for the gem polisher included one-fourth inch steel nuts, glass marbles, wood blocks, and screened gravel. The quality of work is good but the process is slow.

Tests are now being made in the laboratory with horizontal plate planters and polished seed screened between 1/64 inch size limits. The seed plate cell diameter and plate thickness are matched to the seed size so that more accurate cell fill and seed distribution can be attained. Table 2 shows the results of cell and seed sizes and the resulting cell fill for each combination.

Planter manufacturers have indicated that seed plates with cell sizes based on the results of these tests will be made available to growers for next season's planting if plates can be manufactured in time.

Present results would indicate that new designs and refinements in planters combined with a truly satisfactory method of polishing seed will result in improved germination stands and contribute materially to complete mechanization of sugar beet production.

Literature Cited

(1) Barmington, R. D. 1950. Physical factors of the soil affecting beet seedling emergence. Proc. Am. Soc. of Sugar Beet Technol. VI: 228-233.

(2) Barmington, R. D. 1956. Metering and test results of some foreign and domestic sugar beet planters. J. Am. Soc. Sugar Beet Technol. IX (1): 44-50.

A Preliminary Study of the Relationship of the Sugar Beet Nematode Heterodera Schachtii, to Three Varieties of Red Tomatoes

JOSEPH NEMAZI[1]

Received for publication July 13, 1961

The most practical method of controlling the sugar beet nematode, *Heterodera schachtii* Schmidt, has been accomplished primarily by growing a non-host crop for at least four years between sugar beet crops.

Tomato, *Lycopersicum esculentum*, was considered by Thorne (4) to be a non-host plant and he recommended that it be grown in crop rotation for fields infested with *H. schachtii*. Raski (3) found two yellow varieties of tomatoes, Golden Queen and Jubilee, to be hosts of *H. schachtii* but he did not consider Earliana, a red tomato variety, to be a host. Golden and Shafer (1) reported that Pearson 40, a red variety of tomato was susceptible to *H. schachtii* with an infection-index rating which varied from light (2.0) to heavy (4.0) with an average index rating of 2.1. The sugar beet was indexed as heavy (4.0).

An experiment was conducted by the Utah-Idaho Sugar Company at West Jordan, Utah, to determine if the tomato varieties grown locally were hosts to *H. schachtii* and to investigate the cause of variation in infection as reported by Golden and Shafer (1) and Raski (3).

Fifteen plants from each of three red tomato varieties, Hybrid T3, Moscow A, and Moscow B (commercial) were planted March 15, 1960 in a field heavily infested with *H. schachtii*. After 70 days from the date of planting, tomato roots were dug and washed. All three varieties tested were found to be lightly infested with nematode cyst. These cysts were identified as *H. schachtii* after microscopic examination of the cysts by the author. E. C. Jorgensen[2], USDA nematologist, concurred in this identification.

Of the three varieties tested, the smallest number of female larvae were found to be feeding on the rots of the Hybrid T3. Upon microscopic examination of the roots it was found that the three varieties apparently varied in their resistance to *H. schachtii* since a few of the third stage female larvae were imbedded under the epidermis of the tomato roots and were unable to break

[1] Research Agronomist, Utah-Idaho Sugar Company, West Jordan, Utah.
[2] Personal communication.

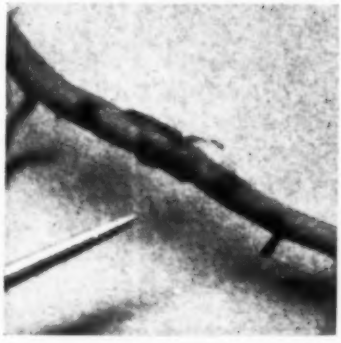

Figure 1.—Brown third stage female larvae of *Heterodera schachtii* imbedded in a tomato root.

through to complete their life cycle (Figure 1). The females trapped inside the roots were brown in color and were found to be nearly devoid of eggs and larvae. Approximately 50% more females were trapped in the roots of Hybrid T3 than in those of the other two varieties. The mechanism of trapping has been reported by Hijner (2) in *Beta patellaris*, where the larvae of *H. schachtii* penetrated into the roots of *B. patellaris*, but were incapable of developing and completing their life cycle.

During the study of the tomato roots, it was observed that secondary roots with thicker epidermis had entrapped more female nematodes than the smaller roots. The Hybrid T3 plants had more vigorous roots and thus fewer nematodes were able to break through the roots and complete their life cycle as compared to the other two varieties.

The apparent difference in resistance to *H. schachtii* exhibited by the three tomato varieties tested has strengthened the contention of Golden and Shafer (1) that resistance exists in some red tomato varieties.

This study points to the fact that varietal resistance may be due to genetic variation in the tendency of tomato roots to trap the female larvae of *H. schachtii*. However, as stated, the three varieties tested had light infection which may rule out the possibility of using these tomato varietes as a trap crop.

Nematologists and tomato breeders should be encouraged to select and breed tomato varieties completely resistant to *H. schachtii* so that tomatoes could be grown in crop rotations in sugar beet districts without any danger of increasing the nematode infestation.

In some areas where *H. schachtii* infestation is widespread it is desirable that the rotation period between beet crops be shortened. A possible solution would be the development of a tomato variety that causes hatching of nematode larvae in large numbers with a subsequent trapping of the adult female within the tomato root.

Literature Cited

(1) GOLDEN, A. M., and SHAFER, THELMA. 1959. Susceptibility of tomato (*Lycopersicum esculentum*) to the sugar beet nematode (*Heterodera schachtii*). Plant Disease Reporter. 43: 1196-1197.

(2) HIJNER, J. A. 1951. Combating nematodes with wild beets. Mededeelingen Institute Rationale Suikerproductie. 21: 1-15.

(3) RASKI, D. J. 1952. On the host range of the sugar beet nematode in California. Plant Disease Reporter. 35: 5-7.

(4) THORNE, G. 1952. Control of the sugar beet nematode. U. S. Dept. Agr. Farmers Bull. 2054, p. 12.

An Evaluation of Insecticides for Control of the Sugar Beet Root Maggot in Southern Alberta[1]

A. M. HARPER[2], C. E. LILLY[2], AND P. BERGEN[3]

Received for publication January 23, 1961

The sugar beet root maggot, *Tetanops myopaeformis* (von Roeder), is a pest of sugar beets in the light sandy soil areas of southern Alberta. Approximately 10,000 acres in this area are treated annually with insecticides for control of this insect.

The treatment now recommended in southern Alberta is heptachlor impregnated in ammonium phosphate fertilizer (11-48-0) and placed in the drill row at the time of seeding. This recommendation stems from an experiment (4)[4] carried out in 1956, the results of which are as follows:

Treatment (2 lb. toxicant per acre)	Number of beets		Damage rating	Beet yield (lb)
	Before thinning	After thinning		
Heptachlor	2045.5*	450.5*	1.53*	2291.5*
Dieldrin	2230.3*	474.8*	1.93	1983.5
Untreated	2794.0	514.8	2.05	1963.3
Aldrin	1948.5*	441.0*	2.07	1829.5
L.S.D. (.05)	386.5	38.2	0.27	223.1

Because heptachlor was phytotoxic when placed in the drill row, both in the above test and in commercial plantings, and because of the possibility of development of strains of *T. myopaeformis* resistant to heptachlor, other insecticides were tested. This paper compares heptachlor with three other insecticides, Dylox[5], Guthion[6], and Di-syston[7], for maggot control and phytotoxicity.

Materials and Methods

The insecticides were tested in irrigated sugar beet fields at Barnwell and Fincastle, Alberta, approximately 30 and 40 miles east of Lethbridge. Sugar beets had been grown in both fields

[1] Contribution from the Entomology Section, Research Station, Canada Department of Agriculture, Lethbridge, Alberta.

[2] Entomologist, Research Station, Lethbridge.

[3] Plant breeder, Canadian Sugar Factories Limited, Taber, Alberta.

[4] Numbers in parentheses refer to literature cited.

[5] Bayer L13/59; active ingredient is dimethyl-(1-hydroxy-2, 2, 2-trichloroethyl) phosphonate.

[6] Bayer 17147; 0, 0-dimethyl S-(4-oxo-1, 2, 3-benzotriazinyl-3-methyl) phosphorodithioate.

[7] Bayer 19639; 0, 0-diethyl S-2-(ethylthio) ethyl phosphorodithioate.

the previous year and both stands had been severely infested with maggots. The soil at both locations was Chin fine sandy loam, the pH being 8.0 at Barnwell and 7.25 at Fincastle. Plots were set out at each site in a randomized block of four replications. Each plot was 60 feet long and eight rows wide.

At the time of seeding the field was fertilized with 100 pounds per acre each of ammonium phosphate (11-48-0) and ammonium nitrate (33-0-0). The fertilizer was placed 2 inches to one side of the drill row and at a depth of 5 inches. The seed beds in both fields were relatively dry at time of seeding. The beets were seeded on May 13 and harvested on October 1 at Barnwell and seeded on May 14 and harvested on October 14 at Fincastle.

The following treatments were tested: 5% granular heptachlor at 1 pound of toxicant per acre; 5% granular Guthion at 1 pound of toxicant per acre; 5% granular Di-syston at 1 pound of toxicant per acre; and Dylox (50% wettable powder) at 4½ pounds of toxicant per acre.

A shoe drill was used to seed all the plots and to apply the first three insecticides in the drill rows with the seed. Dylox was sprayed on the plants and soil surface on June 11 and 20 and July 2 for control of the flies. This insecticide was applied on each date at 1.5 pounds per acre with a low-pressure, tractor-mounted boom sprayer that delivered 13 gallons of liquid per acre. All plots were sprayed with toxaphene on July 3 for control of larvae of the sugar beet webworm.

The stands before thinning were determined on June 15 by counting the "beet-containing" inches in 100 inches of each of the four central rows of all plots. The stands after thinning were determined on July 2 by counting all beets in the four central rows. At harvest the beets in 50 feet of each of the two central rows were counted, rated for maggot damage, and used to determine percentages of beets infested. Damage ratings ranged from 0 for undamaged to 3 for severely damaged beets.

The number of maggots surviving each treatment was estimated by counting those found in the soil around two beets selected at random in each plot; each soil sample was 1 square foot in size and centered on a beet. At Barnwell, samples were taken to a depth of 16 inches; because most of the maggots at that location were in the upper 10 inches of soil, the samples at Fincastle were taken only to that depth. Sampling was done two weeks earlier at Barnwell than at Fincastle.

Table 1.—Mean stands of sugar beets in plots treated with various insecticides to control the sugar beet root maggot, 1959.

	Stand before thinning		Stand after thinning		Stand at harvest	
Location	Treatment	Beet-containing inches[a]	Treatment	No. of beets[b]	Treatment	No. of beets[c]
Barnwell	Dylox	50 [d]	Dylox	210	Dylox	95
	Guthion	46	Guthion	191	Guthion	89
	Check	42	Check	186	Check	89
	Heptachlor	34	Heptachlor	179	Heptachlor	86
	Di-syston	9	Di-syston	80	Di-syston	44
Fincastle	Guthion	71	Guthion	218	Heptachlor	93
	Heptachlor	63	Check	217	Guthion	90
	Check	63	Heptachlor	214	Dylox	79
	Dylox	62	Dylox	206	Check	78
	Di-syston	22	Di-syston	135	Di-syston	60

[a] Beet-containing inches in 400 inches.
[b] Beets in 240 row-feet.
[c] Beets in 100 row-feet.
[d] Means connected by the same vertical line are not significantly different at P = 0.05.

Yield was determined by weighing all beets in 50 feet of each of the two central rows of all plots. These beets were then sampled with a multi-saw rasp for sugar determination. Sugar per acre was determined by multiplying yield of beets by percentage sugar.

All the data were compared at the 5% level of significance by means of Duncan's (3) multiple range test.

Results and Discussion

Stands

At both locations the Di-syston-treated plots had significantly lowered stands on all sampling dates (Table 1). At Barnwell, other treated plots were not significaantly different from the check, but at Fincastle heptachlor plots had a significantly higher stand at harvest than did the other plots except those treated with Guthion. These differences in stand were caused by phytotoxicity in the Di-syston plots and probably by maggots killing beets in the check and Dylox plots.

Damage

Beets from heptachlor-treated plots had a significantly lower damage rating at both locations than did those from the check and Dylox-treated plots (Table 2). At Barnwell, beets from plots treated with Di-syston and Guthion had significantly less damage than did those in the check plots.

Table 2.—Damage by the sugar beet root maggot in plots treated with various insecticides, 1959.

Location	Treatment	Damage rating	Treatment	Percentage of beets infested
Barnwell	Check	1.34	Check	79
	Dylox	1.16	Dylox	77
	Di-syston	0.79	Di-syston	61
	Guthion	0.72	Guthion	56
	Heptachlor	0.33	Heptachlor	31
Fincastle	Check	1.77	Dylox	93
	Dylox	1.76	Check	91
	Guthion	1.40	Guthion	86
	Di-syston	1.37	Di-syston	82
	Heptachlor	1.06	Heptachlor	71

[a] Means connected by the same vertical line are not significantly different at P = 0.05.

At Barnwell the percentage of infested beets from the heptachlor-treated plots was significantly lower than for the other plots. Guthion-treated plots had a significantly lower percentage of infested beets than did the Dylox-treated and the check plots. At Fincastle the means for percentages of infested beets were not significantly different but exhibited the same trend as those at Barnwell (Table 2).

At both locations heptachlor-treated plots had the lowest damage rating and the smallest percentage of infested beets. Guthion- and Di-syston-treated plots were intermediate in those respects, while the check and the Dylox-treated plots showed the highest damage ratings and highest percentages of infested beets. Guthion may be suitable as an alternative to heptachlor if the maggot becomes resistant to the latter insecticide.

Number of maggots

At Barnwell the number of maggots in the heptachlor-treated plots was significantly lower than in the Dylox-treated and check plots but not significantly lower than in those treated with Guthion and Di-syston. The mean number of maggots per beet was 4 for heptachlor, 16 for Guthion, 18 for Di-syston, 32 for Dylox, and 37 for the check. There was no significant difference in numbers of maggots in the plots at Fincastle. The number of maggots varied directly with the damage at Barnwell but not at Fincastle. The samples at Barnwell were examined for maggots by three pairs of workers during favorable weather while those at Fincastle were examined by eight people working individually during adverse weather. These factors may have

accounted for the lack of significance between counts taken at Fincastle.

Yields

At Barnwell sugar beet yield, percentage sugar, and amount of sugar per acre from the Di-syston-treated plots were significantly lower than those from the other plots (Table 3). At Fincastle the yields and percentages of sugar were again lower on the Di-syston plots but not significantly lower. Reduction in stands at both locations due to phytotoxicity from Di-syston resulted in larger beets with lower sugar content.

A significant increase in yield was not obtained with the use of heptachlor, although root damage was significantly reduced. Contrary to observations in 1955 and 1956, stands on untreated plots were not noticeably reduced as a result of maggot damage. This seems to indicate that in the irrigated areas, where moisture is not a limiting factor and where high soil fertility can be maintained, significant reductions in yield may not occur if maggot feeding does not seriously reduce stands. In southern Alberta it has often been observed that where the tap root has been severely damaged or severed, plants have recovered by putting out numerous secondary roots when moisture was adequate (Figure 1). Beets grown on dry land, however, where moisture may be a limiting factor, are probably more adversely affected by attacks of the maggot and their recovery is less likely; this may indicate why application of heptachlor in Manitoba and North Dakota (1, 2) generally results in greater yield increases than in Alberta.

Table 3.—Yields of beets and sugar from plots treated with various insecticides to control the sugar beet root maggot, 1959.

Location	Treatment	Beets (tons/acre)	[a]	Treatment	Percentage sugar	Treatment	Sugar (lb/acre)
Barnwell	Dylox	14.3		Dylox	17.5	Dylox	5020
	Guthion	13.5		Check	17.3	Guthion	4600
	Heptachlor	13.4		Heptachlor	17.0	Heptachlor	4580
	Check	12.9		Guthion	17.0	Check	4460
	Di-syston	9.7		Di-syston	15.3	Di-syston	2960
Fincastle	Heptachlor	13.4		Dylox	16.7	Heptachlor	4420
	Dylox	13.0		Heptachlor	16.6	Dylox	4340
	Guthion	12.9		Cuthion	16.6	Guthion	4280
	Check	12.1		Check	15.9	Check	3840
	Di-syston	11.6		Di-syston	15.8	Di-syston	3660

[a] Means connected by the same vertical line are not significantly different at $P = 0.05$.

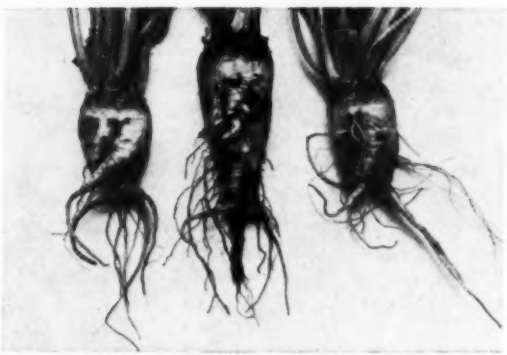

Figure 1.—Sugar beets that were damaged by sugar beet root maggots early in the season. These beets put out numerous secondary roots.

Summary

Heptachlor placed in the drill row at 1 pound of toxicant per acre gave the best control of the sugar beet root maggot. Guthion gave a measure of control and did not cause significant phytotoxicity. Di-syston also gave control of maggots but was too phytotoxic to be recommended in southern Alberta. Three sprays of Dylox for control of the flies did not significantly reduce subsequent larval populations and the resulting damage to the plants.

Literature Cited

(1) ALLEN, W. R., ASKEW, W. L., and SCHREIBER, K. 1959. Relation of sugar-beet root maggot control by insecticides to sugar-beet yield. J. Am. Soc. Sugar Beet Technol. 10: 330-334.

(2) CALLENBACH, J. A., GOJMERAC, W. L., and OGDEN, D. B. 1957. The sugar beet root maggot in North Dakota. J. Am. Soc. Sugar Beet Technol. 9: 300-304.

(3) DUNCAN, D. B. 1955. Multiple range and multiple F tests. Biometrics 11: 1-42.

(4) HARPER, A. M., LILLY, C. E., MCDONALD, S., SWAILES, G. E., and STORY, T. P. Unpublished. Chemical control of sugar-beet root maggot, *Tetanops myopaeformis* (von Roeder). Technical Report for 1956. Crop Insect Section, Science Service Laboratory, Lethbridge, Alberta.

A Comparison of Methods of Applying Heptachlor for Control of the Sugar Beet Root Maggot in Southern Alberta[1]

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The sugar beet root maggot, *Tetanops myopaeformis* (von Roeder), was a problem in the light sandy soil areas near Taber, Alberta, from 1934 to 1937 and from 1955 to 1959. The maggot has also been a problem in the following areas of western North America in recent years: North Dakota (2)[4], Idaho (6), and Manitoba (1).

For control of this pest the authors (5) have recommended that heptachlor-impregnated fertilizer be placed in the drill row at time of seeding. As the degree of phytotoxicity and control of the maggot varied with the different seed drills used (7) the following experiment was designed to determine whether the placement of the treated fertilizer was the major factor causing this variation. Because most of the new drills do not have fertilizer attachments and because heptachlor-impregnated fertilizer had shown some phytotoxicity, other methods of applying heptachlor were tested.

Materials and Methods

The experiment was conducted on irrigated fields at two locations, Barnwell and Fincastle, Alberta, 30 and 40 miles east of Lethbridge. Sugar beets had been grown in both fields in the previous year and both stands had been severely infested with maggots. The soil was Chin fine sandy loam at both locations, the pH being 8.0 at Barnwell and 7.25 at Fincastle. At time of seeding, the field was fertilized with 100 pounds per acre each of ammonium phosphate (11-48-0) and ammonium nitrate (33-0-0). The fertilizer was placed 2 inches to one side of the drill row and at a depth of 5 inches. The soil at both locations was relatively dry at time of seeding.

The experiment was set out in a randomized block design with plots 60 feet long and four drill-rows wide. The following ten treatments were replicated six times at each location:

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[1] Contribution from the Entomology Section, Research Station, Canada Department of Agriculture, Lethbridge, Alberta.

[2] Entomologist, Research Station, Lethbridge.

[3] Plant breeder, Canadian Sugar Factories Limited, Taber, Alberta.

[4] Numbers in parentheses refer to literature cited.

Treatment	Rate of toxicant	Seed drill
1. Granular heptachlor	1 lb per acre	Disc
2. Heptachlor impregnated in 11-48-0	1 lb per acre	Disc
3. Check	Untreated	Disc
4. Granular heptachlor	1 lb per acre	Shoe
5. Heptachlor impregnated in 11-48-0	1 lb per acre	Shoe
6. Check	Untreated	Shoe
7. Heptachlor emulsion (5-inch band)	2½ lb per acre	Shoe
8. Granular heptachlor (5-inch band)	2½ lb per acre	Shoe
9. Granular heptachlor (5-inch band)	5 lb per acre	Shoe
10. Seed treatment—wettable powder with methyl cellulose sticker	2 oz per acre	Shoe

The disc seed drill used for treatments 1 and 2 placed the heptachlor-impregnated fertilizer and the granular heptachlor approximately 2 inches to one side of the drill row while the shoe drill used in treatments 4 and 5 placed the chemicals in the drill row with the seed. The treated seed was sown at 6 pounds per acre from a "v"-belt rather than through the regular seed-metering attachment.

Plots were seeded at Barnwell on May 13 and harvested on October 1 and were seeded at Fincastle on May 14 and harvested on October 14. All plots were sprayed with toxaphene on July 3 for control of the sugar-beet webworm, *Loxostege sticticalis* L.

The stands prior to thinning were determined on June 15 by counting the "beet-containing" inches in 100 inches of each of the two central rows in the plots. The stands after thinning were determined on July 2 by counting all the beets in the two central rows. At harvest all the beets in 50 feet of each of the two central rows were counted and rated from 0 to 3 for maggot damage. Rating 0 was for undamaged beets and 3 for those severely damaged. Percentage infestation was obtained by determining the percentage of beets in every plot that showed maggot damage.

Yield was determined by weighing all beets in the two central rows of each plot. The beets were then sampled with a multi-raw rasp for sugar determination. Sugar per acre was obtained by multiplying beet yield by percentage sugar.

All the data were compared at the 5% level of significance by Duncan's (3) multiple range test.

Table 1.—Mean stands of sugar beets in plots treated with heptachlor to control the sugar beet root maggot, Barnwell, Alberta, 1959.

Stand before thinning			Stand after thinning		Stand at harvest	
Treatment	No. of beet-containing inches[a]		Treatment	No. of beets in 120 row-feet	Treatment	No. of beets in 100 row-feet
Seed treatment	55		Band spray	123	Band spray	106
Band 2½ lb H	37	[b]	Seed treatment	122	Seed treatment	106
Check (S)	37		Check (S)	121	Check (S)	99
Band spray	36		Band 2½ lb H	113	Band 2½ lb H	94
Check (D)	35		Check (D)	112	Check (D)	91
Heptachlor 1 lb (S)[c]	34		11-48-0 + H (D)	112	Heptachlor 1 lb (S)	90
Heptachlor 1 lb (D)[d]	33		Heptachlor 1 lb (D)	110	Heptachlor 1 lb (D)	89
11-48-0 + H (D)	31		Heptachlor 1 lb (S)	110	11-48-0 + H (D)	87
Band 5 lb H	20		Band 5 lb H	101	Band 5 lb H	83
11-48-0 + H (S)	16		11-48-0 + H (S)	80	11-48-0 + H (S)	65

[a] "Beet-containing" inches in 200 inches.
[b] Means connected by the same vertical line are not significantly different at P = .05.
[c] (S)—Seeded with a shoe drill that placed the heptachlor and heptachlor-impregnated fertilizer in the drill row.
[d] (D)—Seeded with a disc drill that placed the heptachlor and heptachlor-impregnated fertilizer two inches to one side of the drill row.

Table 2.—Mean stands of sugar beets in plots treated with heptachlor to control the sugar beet root maggot, Fincastle, Alberta, 1959.

Stand before thinning			Stand after thinning		Stand at harvest	
Treatment	No. of beet-containing inches[a]		Treatment	No. of beets in 120 row-feet	Treatment	No. of beets in 100 row-feet
Seed treatment	61		11-48-0 + H (D)	127	Seed treatment	106
11-48-0 + H (D)[c]	14	[b]	Seed treatment	124	11-48-0 + H (D)	98
Band spray	35		Heptachlor 1 lb (D)	122	Band spray	93
Heptachlor 1 lb (D)	29		Check (D)	122	Heptachlor 1 lb (D)	92
Check (D)	26		Band spray	120	Check (D)	90
Check (S)	25		Check (S)	107	Band 2½ lb	79
Band 2½ lb H	22		Band 2½ lb H	100	Heptachlor 1 lb (S)	78
Heptachlor 1 lb (S)[d]	19		Heptachlor 1 lb (S)	99	Check (S)	73
Band 5 lb H	16		Band 5 lb H	84	Band 5 lb H	59
11-48-0 + H (S)	15		11-48-0 + H (S)	75	11-48-0 + H (S)	49

[a] "Beet-containing" inches in 200 inches.
[b] Means connected by the same vertical line are not significantly different at P = .05.
[c] (D)—Seeded with a disc drill that placed the heptachlor and heptachlor-impregnated fertilizer two inches to one side of the drill row.
[d] (S)—Seeded with a shoe drill that placed the heptachlor and heptachlor-impregnated fertilizer in the drill row.

Results and Discussion

Stands

Mean stand counts prior to thinning, after thinning, and at harvest are shown in Tables 1 and 2.

Where the disc drill was used there were no differences in stands except in the counts made prior to thinning at Fincastle. There the stands in plots treated with heptachlor-fertilizer were better than those of the check and those treated with granular heptachlor. These results indicate that when the heptachlor-impregnated fertilizer and the heptachlor were placed 2 inches from the seed there was no phytotoxicity.

Where the shoe drill was used the heptachlor-fertilizer reduced stands. An examination of the mean stands prior to thinning and after thinning indicates that the fertilizer in the drill row was the main factor contributing to phytotoxicity. This agrees with work done by Dubetz (4), who found that nitrogen fertilizers caused phytotoxicity when the soil was dry at time of seeding.

At both locations the plots treated with the heptachlor spray and those seeded with the treated seed produced better stands than those treated with the heptachlor-fertilizer or with 5 pounds of granular heptachlor. At Fincastle the former two treatments also had stands after thinning and at harvest that were higher than those of the check plots seeded with the shoe drill. The 5-pound granular heptachlor treatment resulted in stands at Barnwell before thinning, after thinning, and at harvest that were lower than those of the check plots. The heptachlor-fertilizer treatment resulted in all stands being lower than the check at both locations. Coating the seed with methyl cellulose and heptachlor did not reduce germination. Tests in the greenhouse in May confirmed that there was no difference in emergence between untreated and heptachlor-treated seed.

Maggot damage

The mean damage rating and percentage of beets exhibiting damage at both locations are shown in Tables 3 and 4. There was a significant difference in maggot damage between the two locations, the damage being much lower at Barnwell.

Where the disc drill was used at Barnwell beets grown in plots treated with 1 pound of heptachlor showed less maggot damage than those grown in plots treated with heptachlor-fertilizer or those that were not treated. At Fincastle there were no significant differences but the trend was the same. At Barnwell beets from the check plots had a higher percentage infestation than those from the plots treated with heptachlor-fertilizer,

Table 3.—Damage by the sugar beet root maggot in plots treated with Heptachlor, Barnwell, Alberta, 1959.

Treatment	Damage rating	Treatment	Percentage of beets infested
Check (D)	1.19	Check (D)	80
Check (S)	1.11	Check (S)	71
11-48-0 + H (D)	0.94	11-48-0 + H (D)	65
Heptachlor 1 lb (D)	0.67	Heptachlor 1 lb (D)	50
Band spray	0.42	Seed treatment	36
Seed treatment	0.39	Band spray	33
11-48-0 + H (S)	0.33	11-48-0 + H (S)	30
Heptachlor 1 lb (S)	0.28	Heptachlor 1 lb (S)	25
Band 2½ lb	0.25	Band 5 lb (S)	24
Band 5 lb	0.25	Band 2½ lb	22

[a] Means connected by the same vertical line are not significantly different at P = .05.

Table 4.—Damage by the sugar beet root maggot in plots treated with heptachlor, Fincastle, Alberta, 1959.

Treatment	Damage rating	Treatment	Percentage of beets infested
Check (S)	1.80	Check (D)	90
Check (D)	1.63	Check (S)	88
11-48-0 + H (D)	1.59	11-48-0 + H (S)	87
Band spray	1.40	Band spray	86
Heptachlor 1 lb (S)	1.26	Heptachlor 1 lb (S)	81
Heptachlor 1 lb (D)	1.24	Heptachlor 1 lb (D)	77
11-48-0 + H (S)	1.08	Band 2½ lb H	76
Band 2½ lb	1.04	11-48-0 + H (S)	75
Seed treatment	0.97	Seed treatment	70
Heptachlor 5 lb	0.87	Heptachlor 5 lb	62

[a] Means connected by the same vertical line are not significantly different at P = .05.

which in turn had a higher percentage infestation than those from the heptachlor-treated plots. At Fincastle no significant differences were found but again the trend was the same. The decrease in damage to beets on the plots treated with heptachlor

alone may have been attributable to slight drift of the granular heptachlor into the drill row, whereas the heptachlor-impregnated fertilizer, being heavier, remained to one side of the drill row.

Where the shoe drill was used at Barnwell all chemical treatments reduced the maggot damage to the beets and, also, the percentage of beets exhibiting damage, but there were no differences between chemical treatments. At Fincastle all treatments except the band spray resulted in lower damage ratings than did the check, but only the seed treatment and the 5-pound granular heptachlor treatment resulted in lower percentages of infestation than did the check.

Yields

The beet yields, sugar content of beets, and sugar yields from all plots are summarized in Tables 5 and 6.

There were no significant differences, at either location, in yields of beets or sugar between plots in which the disc drill was used but, at both locations, the plots treated with heptachlor-fertilizer had a greater yield of both beets and sugar than did those treated with heptachlor alone and those left untreated. This might indicate that in spite of the general addition of fertilizer to the plots prior to the experiment the additional fertilizer still increased beet growth.

Table 5.—Yields of beets and sugar from plots treated with heptachlor to control the sugar beet root maggot, Barnwell, Alberta, 1959.

Treatment	Beets (tons/acre) [a]	Treatment	Percentage sugar	Treatment	Sugar (lb/acre)
Band spray	15.8	Check (D)	17.5	Band spray	5420
Band 2½ lb	15.1	Seed treatment	17.3	Band 2½ lb	5060
Heptachlor 1 lb (S)	14.5	Heptachlor 1 lb (D)	17.2	Heptachlor 1 lb (S)	4940
11-48-0 + H (D)	14.5	Check (S)	17.1	11-48-0 + H (D)	4840
Check (S)	13.8	Band spray	17.1	Check (S)	4740
Seed treatment	13.5	Heptachlor 1 lb (S)	17.1	Check (D)	4680
Check (D)	13.4	11-48-0 + H (S)	17.0	Seed treatment	4660
Heptachlor 1 lb (D)	13.4	Band 5 lb	16.9	Heptachlor 1 lb (D)	4620
11-48-0 + H (S)	13.3	Band 2½ lb	16.8	11-48-0 + H (S)	4500
Band 5 lb	13.2	11-48-0 + H (D)	16.7	Band 5 lb	4460

[a] Means connected by the same vertical line are not significantly different at P = .05.

Table 6.—Yields of beets and sugar from plots treated with heptachlor to control the sugar beet root maggot, Fincastle, Alberta, 1959.

Treatment	Beets (tons/acre)	Treatment	Percentage sugar	Treatment	Sugar (lb/acre)
11-48-0 + H (D)	15.8	11-48-0 + H (D)	17.2	11-48-0 + H (D)	5440
Seed treatment	14.9	Band spray	17.1	Band spray	5020
Band spray	14.7	Heptachlor 1 lb (D)	17.0	Heptachlor 1 lb (D)	4960
Heptachlor 1 lb (D)	14.6	Check (D)	16.8	Check (D)	4840
Check (D)	14.4	Band 5 lb	16.5	Seed treatment	4800
Band 2½ lb	14.1	Check (S)	16.5	Band 2½ lb	4540
Heptachlor 1 lb (S)	13.9	11-48-0 + H (S)	16.3	Heptachlor 1 lb (S)	4500
Check (S)	12.5	Heptachlor 1 lb (S)	16.3	Check (S)	4100
11-48-0 + H (S)	12.2	Band 2½ lb	16.1	11-48-0 + H (S)	3980
Band 5 lb	11.6	Seed treatment	16.1	Band 5 lb	3800

[a] Means connected by the same vertical line are not significantly different at P = .05.

At Barnwell there were no differences in yields of beets or sugar where the shoe drill was used. At Fincastle the seed treatment, the band spray, and the 2½-pound band application of granular heptachlor resulted in greater yields of beets than did the check, the heptachlor-fertilizer treatment and 5-pound band treatment. At Fincastle the seed treatment and the band spray resulted in greater yields of sugar than did the check, the heptachlor-fertilizer treatment and 5-pound band treatment. The latter two gave low yields because they had seriously reduced stands and possibly affected the vigor of the remaining plants. The effectiveness of the seed treatment was greater than expected, indicating that heptachlor may be translocated through the plant. There were no significant differences in percentages of sugar between treatments at either location.

In southern Alberta all sugar beets are grown on irrigated land that is fertilized with ammonium nitrate or ammonium phosphate at 100 or more pounds per acre. In fields where fertility is high and moisture adequate, many of the damaged beets are able to recover and there may be no significant decrease in yield. Thus it appears that where maggot populations are low to moderate, control may not be necessary. However, it is almost impossible to accurately predict the level of population one might expect in a field or the amount of damage that may be caused by any population, so it is probably advisable to use a chemical control in areas where the maggot has caused damage in recent years.

In Manitoba and North Dakota the heptachlor-fertilizer applied in the drill row does not appear to cause as much phytotoxicity as in Alberta, for stands after thinning and at harvest were not reduced in those two areas (1, 2). Because of the phytotoxicity of the heptachlor-fertilizer in Alberta it appears that a modification of our recommended treatment is necessary. As the ammonium phosphate (11-48-0) is the main phytotoxic factor, it might be expedient to increase the concentration of heptachlor so that less fertilizer would be placed in the row at time of seeding. It might also be practical to impregnate the heptachlor on triple super-phosphate fertilizer and to broadcast the nitrogen fertilizer prior to seeding. Dubetz (4) showed that triple super-phosphate placed in contact with the seed was less toxic to beet plants than fertilizer containing nitrogen.

When using seed drills without fertilizer attachments or drills that place the fertilizer outside the drill row, control of the maggots can be obtained with a 5-inch band of granular heptachlor spread over the drill row at 2½ pounds per acre or with 1 pound of granular heptachlor placed in the drill row.

Summary

In southern Alberta placement of heptachlor-impregnated fertilizer influenced both phytotoxicity and maggot control. Better control was obtained when the heptachlor-impregnated fertilizer was placed near the seed than when applied 2 inches to one side of the drill row but phytotoxicity also increased. The fertilizer appeared to be the main factor contributing to the phytotoxicity. High rates of granular heptachlor also caused noticeable phytotoxicity. Granular heptachlor spread in a 5-inch band over the drill row at 2½ pounds per acre or placed in the drill row at 1 pound per acre was not phytotoxic and gave control equal to the recommended heptachlor-fertilizer treatment. A seed treatment of heptachlor also seemed promising, indicating that this insecticide may have some systemic action.

Literature Cited

(1) ALLEN, W. R., ASKEW, W. L., and SCHREIBER, K. 1959. Relation of sugar-beet root maggot control by insecticides to sugar beet yield. J. Am. Soc. Sugar Beet Technol. 10: 330-334.

(2) CALLENBACH, J. A., GOJMERAC, W. L., and OGDEN, D. B. 1957. The sugar beet root maggot in North Dakota. J. Am. Soc. Sugar Beet Technol. 9: 300-304.

(3) DUNCAN, D. B. 1955. Multiple range and multiple F tests. Biometrics 11: 1-42.

(4) DUBETZ, S. 1958. The effect of fertilizers and osmotic pressure on germination. J. Am. Soc. Sugar Beet Technol. 10: 212-219.

(5) HARPER, A. M., LILLY, C. E. MCDONALD, S., SWAILES, G. E., and STORY, T. P. Unpublished. Chemical control of the sugar-beet root maggot, *Tetanops myopaeformis* (von Roeder). Technical Report for 1956. Crop Insect Section, Science Service Laboratory, Lethbridge, Alberta.

(6) JONES, E. W., DOUGLASS, J. R., PARRISH, C. P., and JENSEN, V. 1952. Experiments on control of the sugar-beet root maggot. Proc. Am. Soc. Sugar Beet Technol. 7: 490-496.

(7) LILLY, C. E., HARPER, A. M., MCDONALD, S., and SWAILES, G. E. Unpublished. Chemical control of the sugar-beet root maggot, *Tetanops myopaeformis* (von Roeder). Technical Report for 1957. Crop Insect Section, Science Service Laboratory, Lethbridge, Alberta.

New Hybrid Sugar Beet Varieties for California

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The discovery of cytoplasmic male sterility in the sugar beet by Owen (4)[3] provided a means of producing commercial hybrid seed. Methods of utilizing male sterility in the production of hybrid varieties have been developed and were described by Tolman and Johnson in 1956 (9). Advantages of hybrid varieties from both the standpoint of higher sugar yields and increased disease resistance have now been well demonstrated.

Fall and winter plantings in California require varieties that combine resistance to both bolting and curly top. Varieties used in the coastal valleys should also be resistant to downy mildew. Five new multigerm hybrids which meet the requirements of the major sugar beet-growing districts of California have been developed. These five hybrids are being released by the U. S. Department of Agriculture for use by the sugar beet growers.

Description of Hybrid Varieties

US H2

The US H2 hybrid variety has the parentage (MS of NB1 $\times$ NB3) $\times$ C663. The female parent is an F_1 hybrid between the male-sterile equivalent of the NB1 inbred and the NB3 inbred. The NB1 inbred (2) is an increase of an S_5 line resistant to bolting and curly top. This inbred is an excellent Type O, which insures that all its offspring from crosses to cytoplasmic male steriles will be completely male sterile. The male-sterile equivalent of NB1 has been produced by crossing NB1 to a cytopasmic male-sterile plant found in US 56 and then making repeated backcrosses to NB1.

The NB3 inbred is a moderately bolting-resistant segregate from the CT9 inbred developed by Owen (5). It possesses excellent curly-top resistance but lacks downy-mildew resistance. The F_1 hybrid between the MS of NB1 and NB3 has proved to be a high-performing female parent with excellent curly-top resistance and moderately good bolting resistance. The plants of this hybrid

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[3] Numbers in parentheses refer to literature cited.

are good seed producers and are all male sterile, which makes the hybrid an excellent female parent for use in producing commercial hybrid varieties.

The pollen parent C663 has been derived from a cross between US 15 and US 22. Selections for curly-top resistance, bolting resistance, and high sucrose content were made in the F_2 and F_3 generations. The curly-top resistance of C663 is similar to that of US 75 (3) and the bolting resistance to that of US 56 (6). C663 yields well but tends to be slightly inferior to US 56 and US 75 in sucrose content.

US H2 has been thoroughly tested during the past three years in each of the major sugar beet-producing districts of California. Gross sugar yields and sucrose contents expressed in percent of the performance of US 75 are summarized in Tables 1 and 2. These tests have been conducted by the U. S. Department of Agriculture, the University of California, and the four sugar companies operating in California. An average of these 47 California tests shows that the gross sugar yield of US H2 is 118% and the sucrose content 105% of those of US 75.

Production records (8) from 1958-59 commercial plantings made in the Imperial Valley of California by the Union Sugar Division of Consolidated Foods Corporation show that US H2 yielded 28.6 tons of beets per acre with 15.7% sugar on 235 acres. The commercial check grown in the same fields yielded 24.6 tons per acre with 15.8% sugar on 216 acres. The commercial check used in these fields was US 75 and US 56.

Tests at Jerome, Idaho, showed that US H2 is superior to US 75 in curly-top resistance. The bolting resistance of US H2 is slightly inferior to that of US 56 and US 75. The downy-mildew resistance is poor, and for that reason the variety should not be used near the coast.

US H2 is recommended for fall plantings in the Imperial Valley and for spring plantings in the Central Valley of California. It does not possess sufficient bolting resistance for fall plantings in the Central Valley. It can be used for winter and spring plantings in the inland portions of the coastal valleys, where downy mildew is not a problem.

US H3

The US H3 hybrid variety has the parentage (MS of NB1 × NB3) × C586. It has the same female parent as US H2, but a selection from the US 35 variety has been used as pollen parent in place of C663. This selection has been designated C586 and is the product of three successive selections for bolting resistance. C586 is similar to US 75 in bolting and curly-top resistance.

Table 1.—Gross sugar yields of US hybrid varieties in 1956-59 California variety tests expressed as percent of the yield of US 75.

Variety and year	Coastal District		Central Valley District		Imperial Valley District	
	No. of tests	Percent of US 75	No. of tests	Percent of US 75	No. of tests	Percent of US 75
US H2						
1957	3	113	4	123	3	118
1958	5	111	3	104	7	124
1959	2	124	12	121	8	118
US H3						
1956	1	116	2	112	1	104
1957	4	107	4	101	2	108
1958	5	96	1	96	7	106
1959	2	111	7	104	7	111
US H4						
1956	2	106	2	113	1	108
1957	4	109	3	113	1	113
1958	5	113	1	101	6	105
1959	1	107	11	111	6	115
US H5A						
1958	8	113	1	103	7	115
1959	5	108	0	.....	1	114
US H5B						
1959	8	116	2	126	2	119

Like the parent US 35 variety, it has a relatively high sucrose content but tends to be low in yielding ability. Hybrid varieties which result from the use of C586 as the pollen parent are superior in sucrose content to those in which C663 is utilized as the pollen parent but are inferior in yield.

The results of 43 tests conducted with US H3 during the past four years in the major sugar beet-production districts of California are summarized in Tables 1 and 2. In these tests both the gross sugar yield and the sucrose content of US H3 averaged 105% of US 75. Tests at Salinas, California, and Jerome, Idaho, showed that resistances to curly top, bolting, and downy mildew are similar in US H2 and US H3.

US H3 is adapted to the same districts and planting dates as is US H2. Although sucrose percentages of US H3 are consistently higher, gross sugar yields are lower than those of US H2. US H3 is recommended for areas which have a problem with low sugar.

US H4

The US H4 hybrid variety has the parentage (MS of NB1 × NB2) × C586. The parentage is similar to that of US H3 except for substitution of the NB2 inbred for NB3. The NB2

inbred has been developed from a cross made at Salt Lake City, Utah, in 1943 between a bolting-resistant clone and a self-fertile line. Following selection work at both Salt Lake City and Salinas, California, a high-performing S_5 line was designated NB2. This inbred possesses good vigor, moderately good bolting resistance, and fair curly-top resistance. It has demonstrated good combining ability especially from the standpoint of sucrose content. The F_1 hybrid between the MS of NB1 and NB2 has moderate resistance to both bolting and curly top. It has good vigor and male sterility and has performed well as a female parent in the production of hybrid varieties.

US H4 possesses moderately good bolting and curly-top resistance. It has been included in 43 variety tests during the past four years. A summary of these tests (Tables 1 and 2) show that it produced an average of 10% more gross sugar per acre than did US 75 and that its sucrose content was 107% of US 75. It has performed especially well in the Imperial Valley where it has maintained a high sucrose content during June and July, when the temperatures are extremely high. The variety may also be used in winter and spring plantings in the coastal valleys of California.

Table 2.—Sucrose contents of US hybrid varieties in 1956-59 California variety tests expressed as percent of the sugar content of US 75.

Variety and year	Coastal District		Central Valley District		Imperial Valley District	
	No. of tests	Percent of US 75	No. of tests	Percent of US 75	No. of tests	Percent of US 75
US H2						
1957	3	104	4	105	3	103
1958	5	102	3	101	7	101
1959	2	102	12	105	8	102
US H3						
1956	1	109	2	109	1	106
1957	4	107	4	106	2	104
1958	5	103	1	107	7	104
1959	2	104	7	107	7	105
US H4						
1956	2	103	2	109	1	107
1957	4	107	3	107	1	111
1958	5	107	1	106	6	105
1959	1	105	11	109	6	108
US H5A						
1958	8	102	1	99	7	100
1959	5	102	0	-----	1	101
US H5B						
1959	8	101	2	99	2	101

US H5A and US H5B

The US H5A hybrid variety has the parentage (MS of NB1 × NB4) × C586 and US H5B has the parentage (MS of NB1 × NB4) × C663. These varieties are similar to US H2 and US H3 except for substitution of the NB4 inbred for NB3. The NB4 inbred is an S_5 line which originated from the same cross as did NB2. It combines bolting and downy-mildew resistance with good combining ability but lacks vigor and is susceptible to curly top. The F_1 hybrid between the MS of NB1 and NB4 has good bolting and moderately good curly-top resistance. Good male sterility and seed-setting ability make this F_1 hybrid a desirable female parent for use in the production of commercial hybrid varieties.

Both US H5A and US H5B possess good bolting and moderately good curly-top resistance. Although accurate evaluations of downy-mildew resistance have not been made, both hybrids are expected to be moderately resistant. The US H5A hybrid was included in 22 variety tests during 1958 and 1959. A summary of these tests (Tables 1 and 2) shows that the gross sugar yield averaged 112% and the sucrose content 101% of US 75. A summary of 12 tests with US H5B in 1959 shows that its gross sugar yield averaged 118% and its sucrose content 101% of those of US 75.

These two hybrids are adapted to fall planting in the Imperial Valley and to winter and spring plantings in the coastal valleys. They lack sufficient curly-top resistance for use in the San Joaquin Valley and in the southern part of the Sacramento Valley.

Seed of the Hybrid Varieties

Seed of the bolting-resistant varieties used in California is produced either in Oregon or in the Tehachapi Mountains of California. Each of the new hybrid varieties reproduces satisfactorily in these two areas provided it is planted by August 15. The hybrid seed is produced either by mixing the pollinator with the male-sterile parent or by the strip method.

The strip method of seed production requires the planting of alternate strips of male sterile and pollinator. For commercial seed production the male-sterile strips are usually 16 rows wide and the pollinator strips four rows wide (Figure 1). The pollinator strips are removed prior to harvest as a precaution against seed mixing. Seed produced by the strip method is entirely hybrid provided pollen-producing plants do not occur in the male-sterile parent and contamination with windborne

foreign pollen is avoided. Production costs are higher with the strip method because more labor is required and approximately one-third of the land is taken up by the pollinator strips.

When a good-performing pollen parent such as C663 or C586 is used the practice of mixing the male-sterile parent and the pollinator has proved to be a satisfactory method of hybrid-seed production. From 5 to 7% of the pollinator is thoroughly mixed with the male-sterile parent prior to planting. The pollinator plants intercross and the seed is harvested along with that of the hybrid. The presence of approximately 5% of C663 or C586 in the commercial hybrid has failed to cause a measurable adverse effect on the performance of the hybrid. This method of seed production has the advantage of bringing the male-sterile plants into close proximity with the pollen source and thereby provides a much higher concentration of pollen around the male-sterile plants (7) than when the strip method of seed production is used. This tends to reduce the amount of out-crossing which occurs when small amounts of pollen are wind-borne from other fields and when occasional pollen-producing contaminants are present in the male-sterile parent. This method of seed production is used extensively with the hybrid varieties described in this paper.

The male-sterile parents used to produce US hybrid varieties have also been made available to the sugar company breeders through the Beet Sugar Development Foundation and are used

Figure 1.—Seed field of the US H2 hybrid sugar beet variety near Salem, Oregon. The male-sterile parent is planted in 16-row strips and the pollen parent in 4-row strips. Photographed June 11, 1959.

in the production of sugar company hybrid varieties. The pollen parents used in these varieties have been developed by the sugar company breeders. Production of hybrid seed (1) in which bolting-resistant US male steriles were used in conjunction with sugar company developed pollinators totalled 918,000 pounds in 1959 and 1,658,000 pounds in 1960.

Summary

Five new sugar beet hybrid varieties adapted to California have been developed.

US H2 and US H3 combine good curly-top resistance with moderately good bolting resistance. US H3 is higher in percent sucrose and lower in gross sugar yield than is US H2. Both varieties are adapted to all beet-growing areas of California with the exceptions of those portions of the coastal valleys subject to downy mildew.

US H4 possesses moderately good bolting and curly-top resistance. Best performance has been in the Imperial Valley where a high sucrose content has been maintained during June and July.

US H5A and US H5B combine good bolting resistance with moderately good curly-top and downy-mildew resistance. They are adapted to the coastal and Imperial valleys.

The male-sterile components of the US hybrid varieties have been made available to the sugar company breeders and are being used extensively in the production of company varieties.

Literature Cited

(1) CAMPBELL, SAM C. 1960. Private communication.

(2) MCFARLANE, J. S. 1954. New non-bolting and mildew-resistant seed releases. Proc. Am. Soc. Sugar Beet Technol. 8 (2): 88-89.

(3) MCFARLANE, J. S. and PRICE, CHARLES. 1952. A new non-bolting curly-top-resistant, sugar beet variety, US 75. Proc. Am. Soc. Sugar Beet Technol. 7: 384-386.

(4) OWEN, F. V. 1945. Cytoplasmic inherited male sterility in sugar beets. J. Agr. Res. 71: 423-440.

(5) OWEN, F. V. 1948. Utilization of male sterility in breeding superior-yielding sugar beets. Proc. Am. Soc. Sugar Beet Technol. 5: 156-161.

(6) PRICE, CHARLES, OWEN, F. V., and CARSNER, EUBANKS. 1948. A new sugar beet variety, US 56, for fall and winter plantings in California. Proc. Am. Soc. Sugar Beet Technol. 5: 181-186.

(7) STEWART, DEWEY and CAMPBELL, SAM C. 1952. The dispersion of pollen in sugar beet seed plots. Proc. Am. Soc. Sugar Beet Technol. 7: 459-469.

(8) STOCK, ALDEN L. 1960. Private communication.

(9) TOLMAN, BION and JOHNSON, RONALD. 1956. Materials and methods used in producing commercial male-sterile hybrids. J. Am. Soc. Sugar Beet Technol. 9: 151-160.

The Composition of Beet Molasses with Particular Reference to Nitrogenous Compounds

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Introduction

In the United States two principal types of beet molasses are produced—straight house and Steffen molasses. Molasses from a straight house is the mother liquor remaining after the economical crystallization of sugar from purified and concentrated sugar beet extracts. In the Steffen process, straight house molasses is treated with calcium oxide to precipitate a calcium complex of sucrose; most of the impurities remain in solution. The precipitate is recycled to the carbonator to recover the sugar and provide a source of lime. Molasses formed after this secondary crystallization of the sugar is Steffen molasses.

A comparative study of Steffen and straight house molasses composition is necessary to determine the most economical use of molasses, as well as being a valuable adjunct to other processing investigations, such as the effect of molasses composition on sugar crystallization. Samples of molasses representative of the various beet processing areas of the United States were furnished by the beet sugar companies and were analyzed at this Laboratory for a number of their chemical constituents. The results of these analyses and additional data obtained in earlier work from this Laboratory are presented in this paper.

Results and Discussion

In Table 1 a comparison is made between Steffen and straight house molasses. High, low, and average values are presented to provide an estimate of the composition extremes as well as the mean values. The averages represent 18 straight house and 13 Steffen samples. The data in the remainder of the tables are from a small number of different samples from various sources. No distinction was made between straight house and Steffen molasses in these tables, since the variation between samples of one kind of molasses is much greater than the difference between the two types.

True purity values (sucrose as a percent of total solids) are presented in Table 1. Sugars, after inversion with invertase,

[1] Western Regional Research Laboratory, Albany, California, a laboratory of the Western Utilization Research and Development Division, Agriculture Research Service, U. S. Department of Agriculture.

Table 1.—Composition of straight house and Steffen beet molasses.

		High	Low	Average
		Percent sucrose on solids		
Purity	Straight house	67.4	59.1	63.2
	Steffen	65.7	61.2	63.4
		Grams/100 g of non-sucrose solids		
Sucrose	Straight house	207.0	144.4	171.4
	Steffen	191.9	157.9	173.1
Reducing substances	Straight house	7.8	1.0	2.0
	Steffen	3.7	0.9	1.6
Raffinose	Straight house	8.4	0.7	3.1
	Steffen	10.9	3.0	6.0
Potassium	Straight house	19.8	11.2	14.4
	Steffen	16.3	8.4	12.0
Sodium	Straight house	6.7	1.4	3.5
	Steffen	6.2	1.4	3.6
Chloride	Straight house	6.6	0.7	3.5
	Steffen	5.8	1.3	3.0
Nitrate	Straight house	3.7	1.1	2.4
	Steffen	3.1	1.3	2.3
Total nitrogen	Straight house	7.0	4.2	6.2
	Steffen	6.5	4.1	5.3
Betaine	Straight house	19.2	10.2	15.4
	Steffen	14.3	9.7	12.6
Amino Acids[1]	Straight house	12.6	6.6	9.6
	Steffen	10.8	6.6	8.5
Amides[2]	Straight house	1.4	0.2	0.8
	Steffen	1.2	0.1	0.7
Anions[3]	Straight house	48.5	40.9	43.4
	Steffen	48.3	38.5	42.9
Anions (Milli-equivalents)[3]	Straight house	573.0	403.0	490.0
	Steffen	497.0	424.0	458.0
Anions (Avg eq. weight)[3]	Straight house	104.5	74.5	89.2
	Steffen	106.1	83.1	93.8

[1] Amino nitrogen times 8.4.
[2] Amide nitrogen as asparagine.
[3] Amphoteric compounds (amino acids, etc.) not included.

were determined chemically by the Munson-Walker method. Solids were determined by vacuum-oven drying. There is little difference in the average purity of the two types of molasses. It must be remembered, however, that more sugar is recovered from the beets by the operation of the Steffen process and less total molasses is produced. Since the ratio of sugar to non-sugar is higher in high purity molasses it is obvious that there is more sugar lost to molasses per unit weight of non-sucrose solids in the high purity samples, although the particular factory concerned may produce less total molasses per ton of beets if it is processing high purity beets. The magnitude of the sugar loss in high purity

molasses is emphasized by the weight of sucrose in molasses per 100 grams of non-sucrose solids (Table 1). The highest purity straight house molasses shows a 20% greater loss of sucrose into molasses than the average. Expressing the results on a tonnage basis, the highest purity molasses carries 34 tons more sugar into molasses for every 100 tons of non-sugar solids than the average. Viewed in a different manner, a factory producing average molasses would bag 34 tons more sugar per 100 tons of non-sucrose solids than the factory producing the highest purity molasses.

There is considerable spread in reducing sugar values in both types of molasses. These differences may be due to variations in processing although the previous history of the beets, including extent of storage and storage temperatures, probably plays an important part.

Since raffinose, when treated with calcium oxide in a solution such as molasses, forms an insoluble calcium complex, Steffen molasses should contain more raffinose than straight house molasses. This is borne out by the results shown in Table 1. However, it is generally accepted that beets grown in the Southern California region contain the minimum and those grown in the Rocky Mountain or Kansas areas contain the maximum raffinose. This is reflected in the molasses; samples from these regions are respectively below and above average in raffinose. A study of the raffinose content of the individual Steffen molasses samples indicates that the amount of raffinose may not always be the controlling factor requiring the discard of molasses, although the decision to discard molasses in factory operations may be based in part on this value.

The two metallic ion impurities present in greatest quantity are potassium and sodium. Steffen molasses contains less potassium than straight house molasses but average about the same sodium content. The proportionate variation in sodium is much greater than for potassium. This may be due, in part, to genetic differences in soil composition, but part of it is due to the addition of sodium carbonate or sodium hydroxide to some factory juices to preserve alkalinity. The addition of sodium carbonate would also account for the similarity of sodium composition in the two types of molasses, because much more sodium carbonate is needed to preserve alkalinity in the Steffen process. Since calcium is precipitated during processing, it is present in much lower quantities, averaging 0.5 and 0.7% on non-sucrose solids for straight house and Steffen molasses, respectively. Magnesium is present in even smaller quantities.

There is a greater variation in chloride content (10 fold) than in any other major component of molasses except raffinose. This variation may be very important. We have some indications that chloride as the sodium or potassium salt is very melassigenic and that a low chloride content is associated with the production of low purity molasses. This finding indicates the desirability of removing chloride and preventing its addition to the factory juices.

A noteworthy feature is that nitrate is nearly as high in Steffen as in straight house molasses. The essence of the Steffen process is the return of sucrose in the form of a sucrose-calcium complex and of other substances forming insoluble calcium compounds to the diffusion juice, while the soluble impurities are filtered off. It would be expected that substances that do not react with calcium and those forming soluble calcium salts, such as nitrate, would appear in lesser quantities in Steffen molasses. This is true for potassium, chloride, and many other substances, but does not appear to hold true for nitrate even though calcium nitrate is very soluble in water.

The average total nitrogen content of Steffen molasses is lower than that of straight house molasses. It is possible that the total weight of nitrogen-containing compounds is also lower, although this does not necessarily follow, since the unidentified nitrogen compounds in Steffen molasses may have a lower percentage nitrogen and hence give a greater weight of compound per unit of nitrogen. The nitrogen compound present in largest amounts in molasses is betaine. Quantitatively, there is a considerable difference between the high and low values of betaine for both types of molasses. Since betaine is not precipitated by calcium ions it passes into the Steffen filtrate and its concentration is less in Steffen molasses. Similar variations are found in the amino acid and amide fractions. The weight of amino acids was calculated from the amino nitrogen content and multiplied by a factor of 8.4 to convert to amino acids. Since glutamine is degraded during processing, leaving asparagine as the principal amide in molasses, total amide nitrogen was calculated as asparagine. The amount of nitrogen present as betaine, amides, and amino acids listed in Table 1 accounts for only 50% of the total nitrogen in molasses.

There are more equivalents of anions present (about 8%) in straight house molasses than in Steffen molasses, but the average equivalent weight of the anions in straight house molasses is about 6% lower. The reason for the lower average equivalent weight of anions is due to lower equivalent weight anions forming more soluble calcium salts and being eliminated in the waste to a greater extent in the Steffen process. The actual

weight of anions in the two molasses is nearly the same, 43 g/100 g of non-sucrose solids, although the composition of the anions differs materially.

Reducing substances, raffinose, potassium, sodium, chloride, nitrate, betaine, amino acids, and amides make up 55% and 51% of the non-sucrose solids of straight house and Steffen molasses, respectively. Since the total for the impurities listed in Table 1 are 4% less for Steffen molasses, some undetermined and possibly unidentified substances appear to be concentrated in Steffen molasses in a manner similar to the build-up of raffinose which is recycled through the factory as the calcium-raffinose complex.

The quantities of some of the principal anions present in molasses are presented in Table 2. It should be noted that total sulfur was calculated as sulfate, although part of the sulfur is present as sulfite or as an integral part of some organic compounds. Pyrrolidone carboxylic acid (PCA) accounts for about one third of the anions listed in Tables 1 and 2.

Table 2.—Some anions present in beet molasses.

Anion	Grams/100 g of non-sucrose solids
Lactate	4.7
Glycolate	1.4
PCA	9.4
Citrate	1.0
Malate	1.9
Sulfate[1]	4.6

[1] Total sulfur as sulfate.

Source: Owens, H. S., Stark, J. B., Goodban, A. E. and Walker, H. G. 1955. Applications of compositional knowledge to beet sugar technology. Agr. Food Chem. 3: 550-553.

Table 3.—Principal amino acids in beet molasses.

Amino acid	Grams/100 g of non-sucrose solids	Amino acid	Grams/100 g of non-sucrose solids
Alanine	0.8	Leucines	1.3
Aspartic	1.5	Serine	0.7
Gamma-aminobutyric	1.3	Threonine	0.6
Glutamic	1.5	Tyrosine	0.7
Glycine	0.4	Valine	0.6

Source: Goodban, A. E., Stark, J. B. and Owens, H. S. 1953. Amino acid content of sugar beet processing juices. Agr. Food Chem. 1: 261-264.

Table 3 shows the approximate quantity of the principal amino acids found in molasses. Other amino acids, such as tryptophane, are present in lesser quantities. The most economically important is glutamic acid which appears in the Steffen filtrate and can be converted to monosodium glutamate. The other amino acids are important, because they improve the value of beet molasses as a feed supplement or as a fermentation medium.

Table 4 gives the approximate quantities of purine, pyrimidines, and nucleosides found in molasses. If an adequate market could be found, some of these compounds might be economically recovered from beet molasses or other by-product liquors.

An arbitrary classification of nitrogen-containing compounds present in sugar beet liquors has been made on the basis of their reaction to ion exchange resins. The scheme of separation is shown in Figure 1. In this example, a dilute molasses solution is passed through a column of cation resin in the hydrogen form. Compounds retained by the resin and the nitrogen in these compounds are classed as basic even though some of them, amino acids in particular, can be adsorbed by an anion exchange resin. The effluent from the column is passed, in turn, through a column of anion resin in the formate-chloride form. The nitrogen present in the compounds adsorbed by the resins is classed as acidic. The portion not adsorbed by either resin is classed as unadsorbed. The relative amounts of these three classes are shown in Table 5. From the data in earlier tables and other work not presented in this paper, it has been found that nearly 50% of the basic nitrogen is present as betaine. Amino acids account for 25%, while amides, ammonia, and purines constitute an additional 9%. Seventeen percent remains to be identified. About 55% of the acidic nitrogen is present in PCA. Nitrate accounts for 22%, amino compounds for 5% and 18% is unidentified. Six percent was unadsorbed and has not been identified.

Table 4.—Principal purines, pyrimidines and nucleosides in beet molasses.

Compound	Grams/100 g of non-sucrose solids	Compound	Grams/100 g of non-sucrose solids
Adenine	150	Uracil	10
Guanine	100	Uridine	200
Hypoxanthine	10	Unknowns	10
Thymidine	15		

Source: Stark, J. B., Jaouni, T., and Bailey, G. 1956. The purines, pyrimidines and nucleosides in beet diffusion juice and molasses. J. Am. Soc. Sugar Beet Technol. 9: 201-206.

Table 5.—Distribution of beet molasses nitrogen.

Class	Percent
Basic	66
Acidic	28
Unadsorbed	6

Source: Stark, J. B., Goodban, A. E. and McCready, R. M. 1959. The fractionation of nitrogen compounds in beet molasses. J. Am. Soc. Sugar Beet Technol. 10: 571-577.

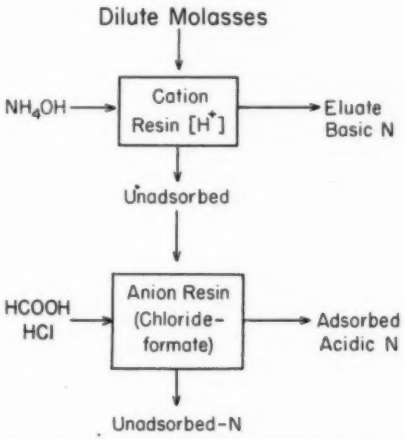

Figure 1.—Fractionation of nitrogen compounds by ion exchange resins, Dowex-50[1] and Dowex-1[1].

Summary

Differences in composition are shown for straight house and Steffen molasses. High, low and average values are shown for purity, sucrose, reducing substances, raffinose sodium, potassium, chloride, total nitrogen ,amino acids, betaine, amides, anions, milliequivalents of anions and average equivalent weight of anions. Raffinose is the only substance determined that is present in significantly greater amounts in Steffen molasses non-sucrose solids than in straight house molasses. Fifty-five percent of the impurities in straight house molasses was determined and only 51% in Steffen molasses. Some unidentified compounds besides raffinose have been concentrated in the molasses during the operation of the Steffen process.

[1] Reference to a company and/or product name by the Department is only for purposes of information and does not imply approval or recommendation of the product to the exclusion of others which may also be suitable.

Two thirds of the nitrogen present in compounds in molasses can be removed on a cation exchange resin. Betaine and various amino acids form most of this fraction. Most of the remaining nitrogen compounds can be adsorbed on a strongly basic anion exchange resin in the formate-chloride form.

An ion exchange procedure is described that can be used to separate a number of nitrogen compounds found in molasses.

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I am indebted to beet sugar companies in all parts of the United States for supplying samples for this study. I wish to thank the following people for analytical determinations: Arthur Bevenue (sulfur and raffinose), Marian Long (amino nitrogen), Harold Lukens (potassium and sodium), Waldo Maher (chloride), Earl Potter (reducing sugars and sucrose) and Henry Wright (amide and total nitrogen).

Evaluation of Telone and D-D in Relation to Planting Time and Fallowing for Control of Sugar Beet Nematode, Heterodera Schachtii Schmidt

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Introduction

During the past few years, experiments with soil fumigants were conducted at the Salt Lake City Nematology Field Station with the objective of developing more effective and less expensive chemical control of the sugar beet nematode. The effects of soil fumigation in relation to early planting and fallowing also were studied. This report summarizes these experiments.

The importance of planting date to beet production in sugar beet-nematode-infested fields was reported by Raski and Lear (1)[2]. Later, Raski and Johnson (2) re-emphasized the importance of early planting. They also reported that maximum activity of sugar beet nematode occurs at temperatures between 70° and 80° F. and that the minimum temperature requirement for the activity of sugar beet nematode larvae is near 70° F. Wallace (3) reported earlier that 77° F. was optimum for emergence of sugar beet nematode larvae, and he also noted that a higher rate of larval emergence occurred under fluctuating temperatures.

Description of Fumigation Experiments

In 1957, an experiment (Table 1) was conducted to compare dosage rates of two commercial nematocides containing 1,3-dichloropropene as the active ingredient. With a standard chisel applicator, the liquid materials were metered through a pressure orifice system and injected 8 inches deep in rows spaced 12 inches apart on a field of Welby fine sandy loam. The surface of the soil was sealed against rapid escape of the chemical by harrowing. The plots were 88 inches (4 rows of beets) wide by 75 feet long. Each treatment was replicated eight times in randomized block design. Date of application was April 17, 1957, and planting date May 3. The two center rows from the middle 25-foot section of each plot were harvested 165 days later.

Another experiment (Table 2) in 1957 in the same field consisted of broadcast treatments applied in strips running the 600-foot length of the field. The treatments were made in triplicate according to a randomized design. Method and time of application, and planting and harvest dates were the same.

[1] Nematologists, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, Salt Lake City, Utah, and Madison, Wisconsin, respectively.

[2] Numbers in parentheses refer to literature cited.

Table 1.—Average yields of sugar beets following soil treatments for control of sugar beet nematode in eight replications, 1957.

Material[3]	Rate per acre		Yield per acre
	Gallons		Tons
Telone[1]	15		22.19
Telone	20		26.61
Telone	25		27.56
D-D[2]	15		19.16
D-D	20		26.51
D-D	25		28.10
None (Control)	0		10.76
LSD 5%		3.28	
LSD 1%		4.37	

Table 2.—Average yields of sugar beets following soil treatments for control of sugar beet nematode in three replications, 1957.

Material	Rate per acre	Yield per acre
	Gallons	Tons
Telone	18	20.86
Telone	25	21.68
D-D	25	19.81
None (Control)	0	9.89

Two additional experiments, completed in 1959, were designed to evaluate the effects of reducing the rates of nematocides when beets were planted early. One of the tests (Table 3) was on a field fallowed during 1958. The treatments were applied in strips 22 feet wide and 600 feet long. Plots were randomized and each treatment was replicated three times. Applications were made in the fall of 1958 to allow the earliest possible planting date. The nematocides were metered through a pressure orifice into the furrow ahead of a plow at a depth of 10 inches. The surface of the soil was sealed against rapid escape of the chemicals by harrowing.

Early plantings were made March 26, 1959, and the late plantings May 22.

Soil and air temperatures were recorded on thermographs placed in the field April 11, 1959.

[1] Furnished by the Dow Chemical Company as a product containing over 90% 1, 3-dichloropropene.

[2] A mixture containing 1, 3-dichloropropene, 1, 2-dichloropropane, and other chlorinated hydrocarbons, furnished by the Shell Chemical Company.

[3] Mention of material and company name is for identification only and does not imply endorsement by U. S. Department of Agriculture.

The near 70° F temperatures reported by Raski and Johnson (1959) to be favorable for sugar beet nematode activity were reached in the plot area by mid-June. Temperatures above 75° were reached shortly thereafter and were sustained until early August. During some years, however, optimum temperatures may be reached earlier as evidenced by records of soil temperatures from a weather station 1½ miles from the test plots.

March planting allowed the beets to grow approximately 75 days before temperatures became optimum for sugar beet nematode activity and 190 days until harvest. Planting May 22, allowed essentially no time after emergence of the beets before favorable temperatures were reached, and only 135 days until harvest.

The second test (Table 4) was conducted in a neighboring field on the same farm. In this instance, there was only one planting date, March 26, 1959, and the field had not been fallowed. Otherwise, except for an additional replicate of each treatment, time, rates, and the method of application of the fumigants and harvest dates were the same.

Table 3.—Average sugar beet yields in date-of-planting and fallow tests from fumigated and unfumigated plots in three replications, 1959.

Material	Rate per acre	Yield per acre		
		Fallowed plots planted March 26	Nonfallowed plots	
			Planted March 26	Planted May 22
	Gallons		Tons	
D-D	15	29.00	25.40	
D-D	25		25.80	16.02
Telone	15	28.17	23.88	
None (Control)	0	27.75	7.42	3.74
LSD 5%		3.59		

Table 4.—Average yields of sugar beets following soil treatment for control of sugar beet nematode in four replications, 1959.

Material	Rate per acre	Yield per acre
	Gallons	Tons
Telone	15	20.29
Telone	20	20.80
Telone	25	21.07
D-D	20	20.78
D-D	25	22.36
None (Control)	0	15.23

Discussion and Conclusions

The plots receiving the 15 gallon-per-acre rates yielded significantly less in 1957 than those receiving 20 or 25 gallons. The plots receiving 25 gallons per acre yielded more than those receiving 20 gallons, but the difference was not significant (Table 1). In 1959, the differences in yield shown in Table 4, between plots receiving reduced rates of fumigant and those receiving normal rates, were not significant. This was probably because of early planting, a lower level of fertility in the plot area, and a lower population level of the sugar beet nematode as evidenced by relatively high yields from the untreated control.

Highest yields were obtained in 1959 from plots which were fallowed the previous season, fumigated, and planted early (Table 3).

Yields from fallowed plots including the check were significantly higher than yields from nonfallowed plots which were otherwise similar. There was no significant difference in yield between treated and untreated fallowed plots, but it is not known whether this result can be consistently duplicated. However, these data suggest that in this experiment fallowing depressed sugar beet nematode populations and can be used under certain conditions as an aid in the control of sugar beet nematode.

Literature Cited

(1) RASKI, D. J. and LEAR, B. 1958. Control of sugar-beet nematode. Calif. Agr. 12: 8,-12.

(2) RASKI, D. J. and JOHNSON, R. T. 1959. Temperature and activity of the sugar-beet nematode as related to sugar-beet production. Nematologica 4 (2) : 136-141.

(3) WALLACE, H. R. 1955. Factors influencing the emergence of larvae from cysts of the beet eelworm, *Heterodera schachtii* Schmidt. J. of Helminthology. 29 (1-2) : 3-16.

Saponin Survey of Various Factories of the 1958 Campaign

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In recent years a great deal of study has been made on floc in beet sugar. The make-up of floc (1, 2)[3] has been characterized and it is generally agreed that saponin is the compound causing the formation of floc. Attempts have been made to eliminate saponin in the factory process by chemical and physical methods (2). Several methods have been devised for the quantitative determination of saponin (3, 4, 5). At the 1958 ASSBT meetings it was reported that successful production of floc-free sugar was obtained by maintaining a high pH in the white pan (6). Examination of operating data for various factories of the American Crystal Sugar Company indicated these findings also to be generally true in regard to high pH of standard liquor and production of floc-free sugar. The scope of this paper was to determine the various factors that contribute to the production of floc-free sugar.

A saponin survey of factory liquors and sugars was initiated, which characterized the pH and saponin content of standard liquor and the saponin concentration of white sugar for various factories. Other liquors and sugars were also analyzed for saponin concentration.

The factories sampled represented four types of operation:

1. Steffen house—batch carbonation
   Factory B
2. Steffen house—continuous carbonation
   Factory C
3. Straight house—batch carbonation
   Factory F
4. Straight house—continuous carbonation
   Factory A
   Factory D
   Factory E

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[3] Numbers in parentheses refer to literature cited.

Table 1.—Factory F, parts per million saponin on dry substance.

Samples	Time (hours)	Saponin on dry substance, ppm							
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Average
Diffusion juice	0	3307	4335	4298	3542	2868	3682	3680	3673
1st carbonation	1	110	101	139	139	86	95	85	108
2nd carbonation	2	110	121	110	128	72	91	123	108
Thick juice	3	120	140	113	136	127	103	101	121
Standard liquor	4	94	97	90	76	85	82	104	90
White sugar	6	1.6	1.9	1.3	1.2	3.0	0.4	1.3	1.5
Intermediate sugar	..	45.6	17.4	26.2	12.5	30.0	22.0	44.0	25.0
Raw sugar	..	120	124	120	112	115	115	108	116
Standard liquor pH		8.8	9.1	8.8	8.0	8.3	8.5	8.4	8.6
Standard liquor purity		93.2	94.3	93.2	93.8	94.2	93.6	93.5	93.7

Table 2.—Factory averages, parts per million on dry substance.

Samples	Time (hours)	Saponin on dry substance, ppm						
		A	B	C	D	E	F	Averages
Diffusion juice	0	3348	2333	1660	2085	2898	3673	2666
1st carbonation	1	109	73	46	111	117	108	94
2nd carbonation	2	116	68	52	91	78	108	86
Thick juice	3	117	71	62	92	93	121	93
Standard liquor	4	91	53	39	55	62	90	64
White sugar	6	6.3	0.8	1.2	0.7	3.4	1.5	2.3
Intermediate sugar	..	30	7.4	3.5	4.2	8.2	25	13.1
Raw sugar	..	77	61	35	82	101	116	79
Standard liquor pH		7.8	8.7	8.1	8.6	8.2	8.6	8.4

A daily survey was made for seven days at the various factories of the following juices and sugars:

1. Diffusion juice
2. First carbonation
3. Second carbonation
4. Thick juice
5. Standard liquor
6. White sugar
7. Intermediate sugar
8. Raw sugar

These were sampled at such a time that the particular juice or sugar sample was representative of the original diffusion juice sample.

The saponin concentrations were determined colorimetrically by the methanol-sulfuric acid procedure (7). All samples were run in duplicate averaged and are reported in ppm saponin on dry substance (D.S.).

Table 1 shows the daily variations for a period of seven days of saponin content of the various liquors and sugars of Factory F. Also listed are the pH values and purities of standard liquor.

Table 2 lists the average saponin content of various liquors and sugars, respectively, of factories A, B, C, D, E, and F.

Discussion of Results

Diffusion juice

Differences existed from factory to factory in saponin content of diffusion juice. The saponin content ranged from 1660 ppm for Factory C to 3673 ppm for Factory F. This variation of saponin content in diffusion juices may be due partly to both location and variety of beets (8). The average saponin content of diffusion juice of all factories was 2666 ppm.

First carbonation

The largest removal of saponin occurs at this station. It is important that optimum operating conditions, including filtration, exist for saponin removal at first carbonation. The straight house factories A, D, E, and F removed an average of 95.7% saponin at first carbonation leaving in solution, in nearly identical amounts, an average of 111 ppm saponin.

The Steffen factories B aand C averaged a 97.1% removal of saponin at first carbonation. An average of 60 ppm saponin remained in solution in first carbonation.

Steffen factories were found to contain approximately half the saponin level found in straight house in first carbonation juice. The authors feel that this may be partly due to the fact

that the diffusion juice of the Steffen factories contained an average of only 66% of the saponin found in the straight house diffusion juice and not because of the effect of the Steffen operation. Mechanical dilution of the saponin concentration of the juice in the first carbonation tank brought about by the addition of sugar from the Steffen house should also contribute to a low saponin content in first carbonation filtered juice.

Although first carbonation serves as the major source of saponin removal, it is not the prime factor in the production of low saponin (floc free) white sugar.

Second carbonation

The results did not indicate that any appreciable amount of saponin was removed by second carbonation.

Thick juice

Thick juice showed approximately the same saponin concentration as found in first and second carbonations.

Standard liquor

It was found that pH and saponin concentration of standard liquor were the controlling factors in the production of white sugar of low saponin content. A striking example of the effect of pH of standard liquor was illustrated by the saponin contents of Factories A and F listed in Table 2. Both factories had practically identical saponin content for like juices and intermediate sugar. Factory F produced a white sugar of 1.5 ppm saponin while Factory A produced a white sugar of 6.3 ppm saponin. The only significant difference between the two factories was the pH of standard liquor, 7.8 for Factory A and 8.6 for Factory F.

Factory C was able to produce white sugar of a saponin content of 1.2 ppm from standard liquor of a pH of 8.1 and saponin content of 39 ppm. This demonstrates that low saponin white sugar may be produced from standard liquor of a low pH if its saponin content is also of a low level. Factory E with a nearly like pH of 8.2 of standard liquor, but with a higher saponin content of 62 ppm in standard liquor yielded white sugar containing 3.4 ppm saponin. Normal standard liquor purities were found to bear no relationship to the saponin content of white sugar.

White sugar

The data of ppm saponin of 48 white sugar samples of all the factories tested were averaged into three groups along with the corresponding pH values and saponin concentrations of standard liquor from which the sugars were produced. The first

Table 3.—Floc groupings based on saponin content of white sugar.

	White sugar ppm saponin	Standard liquor ppm saponin on D.S.	Standard liquor pH
Group 1. (Floc-free sugar)	0.7	56	8.5
Group 2. (Borderline floc-free sugar)	1.3	66	8.5
Group 3. (Floc sugar)	5.0	76	8.0

group consisted of the averages of 17 samples which should be floc-free sugars ranging from 0.4 to 0.9 ppm saponin. The second group consisted of the average of 16 borderline floc-free sugars ranging from 1.1 to 1.9 ppm in white sugar. The third group was made up of the average of 15 samples which should be floc sugars ranging from 2.2 to 7.6 ppm saponin.

Examination of groups 1 and 2 indicated the only difference in standard liquor was a 10 ppm increase of saponin content resulting in a two-fold increase of saponin in white sugar.

Examination of the averages of groups 2 and 3 showed an increase of 10 ppm saponin and a decrease of 0.5 pH units in standard liquor resulting in a four-fold increase of saponin content in white sugar. This indicated the important effect of pH of standard liquor on the production of low saponin content in white sugars.

In Figure 1 the pH of standard liquor was plotted against the saponin content of standard liquor. Examination of Figure 1 points out that with increase in saponin content of standard liquor there must be an increase in pH of standard liquor in order to produce low saponin sugar. Thus by knowing the pH and saponin level of standard liquor it is possible to predict the saponin level of white sugar.

Intermediate sugar

As introduced to the high melter the saponin content of intermediate sugar was low enough to bring about a significant lowering of saponin content from thick juice to standard liquor on a dry substance basis. Factories A and F, which had the highest saponin contents in intermediate sugars, had this lowering effect to a smaller degree.

Raw sugar

Although the saponin content of raw sugar was quite high it did not have a direct effect on the saponin content of standard

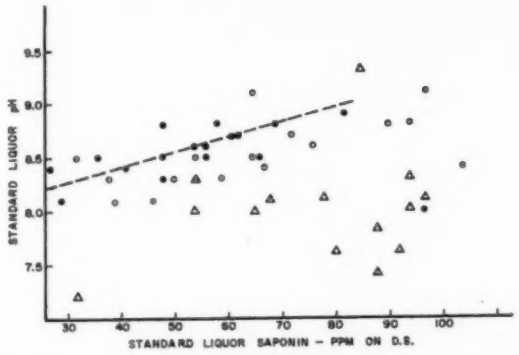

Figure 1.—The effect of the relationship of saponin content and pH of standard liquor on the floc (saponin) content of granulated sugar. The solid dots represent granulated sugars that are floc-free and contain less than 1.0 ppm saponin; the circles represent sugars that are border line floc-free and contain 1.0 to 1.9 ppm saponin; the triangles, sugars, that are floc sugars and contain 2.0 or more ppm saponin.

liquor since it is normally not directly introduced into standard liquor.

Comments on individual factories

Factory A

This factory having a saponin content of standard liquor of 91 ppm, and pH of 7.8 yielded white sugar of 6.3 ppm saponin. It would appear that this factory with such a high saponin level in standard liquor would have to carry a pH of about 8.9 or higher in standard liquor in order to produce floc-free white sugar of less than 1.0 ppm saponin. Another aid would be to obtain greater saponin removal in carbonation if possible.

Factory B

This factory maintained a suitable pH of 8.7 for a saponin content of 53 ppm in standard liquor in order to produce floc-free sugar of 0.8 ppm saponin.

Factory C

Factory C was able to produce sugar of 1.2 ppm saponin from standard liquor of a pH of 8.1 and a saponin content of 39 ppm. A slight increase of saponin content in standard liquor may be enough to cause the formation of floc sugar at an 8.1 pH. It would appear that this factory should strive to maintain at least a slightly higher pH of standard liquor in order to insure production of floc-free sugar.

Factory D

This factory acted similarly to Factory B. Standard liquor had a pH of 8.6, saponin content of 55 ppm yielding saponin content in sugar of 0.7 ppm.

Factory E

This factory did not carry a high enough pH of standard liquor for the amount of saponin present in standard liquor in order to produce floc-free sugar. A pH above 8.5 should yield floc-free sugar.

Factory F

Factory F had a standard liquor of 8.6 pH and 90 ppm of saponin, and yielded white sugar of 1.5 ppm saponin. A higher pH is required for the production of sugar containing less than 1.0 ppm saponin.

Amounts of saponin found in Steffen house operations

The saccharate milk and its components were analyzed for saponin to determine their effect on the total saponin introduced into the factory. Saponin results are found in Table 4.

The amount of saponin entering the factory in saccharate milk was approximately 5% of that entering the house from beets. It appeared that this amount of saponin from Steffen house operations contributed very little to the saponin concentration in carbonation.

Table 4.—Saponin concentrations in Steffen factory operations.

Samples	ppm saponin on D.S.
Cold cake	133
Hot cake	246
Sweet water	85
Saccharate milk	97

Table 5.—Comparison of saponin content of campaign composites of white sugar and pH of standard liquor of 1956, 1957 and 1958 campaigns.

Factory	White sugar saponin ppm			Standard liquor pH values		
	1956	1957	1958	1956	1957	1958
Factory A	6.2	6.3	4.7	7.6	7.6	7.7
Factory B	1.0	0.6	0.7	8.8	9.0	8.9
Factory C	1.9	1.7	1.0	8.0	8.5	8.5
Factory D	3.6	1.4	0.3	7.9	8.0	8.4
Factory E	2.2	2.2	1.8	8.0	8.2	8.3
Factory F	1.0	1.1	1.3	8.8	9.0	8.7
Factory G[1]	1.0	0.9	0.7	7.9	8.0	7.9
Factory H[1]	3.5	4.3	2.1	7.8	7.8	7.9
Factory I	7.0	4.8	2.1	7.6	7.8	8.1
Average	3.04	2.59	1.63	8.04	8.19	8.27

[1] Milk of lime added to high melter.

Comparison of saponin content of campaign composites of white sugar and pH of standard liquor of the 1956, 1957 *and* 1958 *campaigns for nine factories*

These results as pictured in Table 5 demonstrate the beneficial effect of high pH in standard liquor on the production of low saponin sugar.

Summary and Conclusions

pH and saponin content of standard liquor appeared to be the determining factors in the production of white sugar of low saponin (or floc) content. The normal purities as found in standard liquor bore no relationship to the saponin content of white sugar.

The types of carbonation, (batch or continuous) or the types of factory operation, (straight or Steffen) had no bearing on the production of floc-free sugar.

The saponin content of saccharate milk from Steffen operations had no appreciable effect on the saponin content of the house.

The saponin content of diffusion juice varied widely from factory to factory.

Defecation removed 96.5% of the saponin content of diffusion juice of all factories averaged. All of this was done in first carbonation.

Floc-free sugar can be produced when the proper pH is maintained for the saponin level encountered in standard liquor. Thus the saponin level of white sugar can be predicted prior to its actual production when pH and saponin level of standard liquor are known.

Literature Cited

(1) Walker, H. G., Jr. and Owens, H. S. 1953. Beet sugars, acid insoluble constituents in selected samples. J. Agr. Food Chem. 1: 450-453.

(2) Eis, F. G., Clark, L. W., McGinnis, R. A. and Alston, P. W. 1952. Floc in carbonated beverages. Ind. Eng. Chem. 44: 2844-2848.

(3) Walker, H. G., Jr. 1956. Determination of saponins in refined beet sugars. J. Am. Soc. Sugar Beet Technol. 9: 233-237.

(4) West, R. R. and Gaddie, R. S. 1956. Determination of saponin in granulated sugar: Method of H. G. Walker adapted to routine control. J. Am. Soc. Sugar Beet Technol. 9: 146-152.

(5) Johnson, J. R. and Diehl, N. R. 1956. Determination of floc in refined sucrose by coagulation with quarternary amines. J. Am. Soc. Sugar Beet Technol. 9: 221-226.

(6) Gaddie, R. S. and West, R. R. 1958. Relationship between white pan pH and saponin content of the sugar. J. Am. Soc. Sugar Beet Technol. 10: 171-176.

(7) Bauserman, H. M. and Hanzas, P. C. 1957. Colorimetric determination of saponin as found in beet sugars. J. Agr. Food Chem. 5: 773-776.

(8) Olson, R. F. American Crystal Sugar Company. Private communication.

Effect of Nabam Solutions on the Emergence of Larvae from Cysts of Heterodera Schachtii Schmidt.

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Various attempts have been made to develop improved methods for controlling the sugar beet nematode. Studies by Franklin (4)[2] and Raski and Lear (9) suggest that reduction of the expense of chemical control would increase the economic feasibility of this practice. Certain workers maintain that the efficiency of control by crop rotations can be improved by the addition to the soil of a chemical which will induce hatching of larvae from cysts in the soil (1).

Miller and Stoddard (8) reported that disodium ethylene bisdithiocarbamate[3] (Nabam) in water solution retards hatching of eggs of root-knot nematodes (*Meloidogyne* sp.), but that in soil it increased hatching of eggs of both *Meloidogyne* sp. and *Heterodera tabacum* Lownsbery and Lownsbery, 1954. They also reported that better control of root-knot nematodes was obtained by combining Nabam and a nematocide than by either alone. These workers suggested that Nabam or a decomposition product is a "hatching factor" for *Meloidogyne* and *Heterodera*, but no acceleration of hatching of root-knot nematodes resulted when roots and soil were treated with ethylene thiuram monosulphide and ethylene thiuram polysulphide, which were reported by Ludwig and Thorne (7) to be breakdown products of Nabam. No information is available as to the effect of Nabam on hatching of the sugar beet nematode.

Materials and Methods

Two separate tests were conducted. In the first, the hatching effect of Nabam at 500, 1,000, or 2,000 parts per million (ppm) was compared with that of tap water and sugar beet-root diffusate.

The methods used to obtain beet-root diffusate treatment solutions and the conduct of hatching tests were essentially those described by Golden (5) but are briefly reviewed below.

Beet-root diffusate was obtained by intermittently adding tap water to a 4-inch pot containing three vigorously growing sugar

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[2] Numbers in parentheses refer to literature cited.

[3] The formulation of Nabam used by Miller and Stoddard and in the tests reported herein was Dithane D-14 (Rohm and Haas Co.) consisting of 22% Nabam.

Use of trade names and Company names is for identification only and does not imply indorsement by the Department of Agriculture over similar ones not mentioned.

beet (*Beta vulgaris* L.) plants. In this way 200 ml of solution were leached from a single pot in a 24-hour period. Fresh leachate was obtained at weekly intervals throughout the six weeks of the test.

The Nabam solutions for the first test were prepared at the time the test was begun and stored under refrigeration at 1 to 3° C until needed.

The second test (Table 2) included treatments of tap water, beet root diffusate, 1,000 ppm Nabam (solution prepared at the beginning of the test), Maneb (1,000 ppm Nabam plus 1,197 ppm manganese sulfate), Zineb A (1,000 ppm Nabam plus 1,316 ppm zinc sulfate), or Zineb B (1,000 ppm Nabam plus 658

Table 1.—Total numbers of *Heterodera schachtii* larvae emerged from cysts exposed six weeks to various Nabam solutions, tap water, or sugar beet root diffusate.

Treatment	Concentration (ppm)	Replication 1	2	3	4	Total	Average	Percent of hatch in diffusate
Tap water	.....	520	870	540	1,200	3,130	782.5	9.2
Nabam	500	5,240	2,600	2,890	3,840	14,570	3,642.5	42.7
Nabam	1,000	5,150	4,740	6,320	4,920	21,130	5,282.5	61.9
Beet diffusate	.....	9,730	7,960	7,700	8,760	34,150	8,537.5	100.0
Significance							**	
LSD (.05 level)							1,825.0	42.0

Table 2.—Total number of *Heterodera schachtii* larvae emerged from 40 cysts exposed six weeks to various Nabam solutions, tap water, or sugar beet-root diffusate.

Treatment[1]	Replication 1	2	3	4	Total	Average	Percent of hatch in diffusate
Tap water	1,800	2,180	1,990	1,900	7,870	1,967.5	23.0
Maneb	1,810	2,370	2,630	2,110	8,920	2,230.0	26.1
Zineb A	2,390	1,780	2,020	1,520	7,710	1,927.5	22.5
Zineb B	3,880	5,390	3,790	4,090	17,150	4,287.5	50.5
Nabam A	4,980	6,180	4,340	5,190	20,690	5,172.5	60.5
Nabam B	2,830	2,340	2,810	2,470	10,450	2,612.5	30.6
Beet diffusate	10,140	6,820	8,940	8,300	34,200	8,550.0	100.0
Significance						**	
LSD (.05 level)						1,095.0	13.0

[1] Components of treatments:

Maneb—1,000 ppm Nabam + 1,197 ppm manganese sulfate; Zineb A—1,000 ppm Nabam + 1,316 ppm zinc sulfate; Zineb B—1,000 ppm Nabam + 658 ppm zinc sulfate; Nabam A—1,000 ppm Nabam; Nabam B—1,000 ppm Nabam prepared 6 weeks before preparation of all other treatments.

ppm zinc sulfate), and 1,000 ppm Nabam solution prepared six weeks before initiation of the test. Solutions for all other treatments were prepared at the time the test was initiated.

Cysts of *H. schachtii* were removed from soil by washing and screening, transferred to Syracuse watch glasses containing tap water, and refrigerated for 10 days. To insure uniformity of cysts between replications and between treatments, the total mass of cysts was divided into four lots (replications) and each lot subdivided into groups having a number of cysts equal to the number of treatments of the test. The cysts included in a single group were similar in size, shape, color, and plumpness. Individual watch glasses, designated as the first replication of a randomly assigned treatment, continued to receive one cyst from each group until each eventually contained 40 cysts. The process was repeated three more times to give a total of four replications for each treatment.

Approximately 15 ml of each of the treatment solutions were added to individual watch glasses, which were kept in a dark, aerated cabinet in the laboratory during the 6-week test period. At weekly intervals the cysts were transferred to clean watch glasses containing fresh solutions and the emerged larvae preserved in 5% formalin until counted. Aliquot samples from replications which contained large numbers of nematodes were taken to expedite counting. Data for treatments and replications in this randomized complete block experiment were analysed for statistical significance by the "analysis of variance" method.

Results and Discussion

In the first experiment, Nabam at 2,000 ppm inhibited hatching (92 larvae emerged from 160 cysts), whereas hatching in 1,000 ppm was 62%, hatching in 500 ppm was 42%, and hatching in tap water was only 9% of that in beet-root diffusate. Results were erratic and the significance of some of the differences is doubtful.

However, the freshly prepared Nabam solution used in the second experiment gave results very similar to those obtained with a concentration of 1,000 ppm in the first experiment; so it can be concluded that hatching was increased by this material under the conditions of these experiments. The older solution of Nabam gave no better hatches than did tap water, indicating that the hatching effect of Nabam decreases with age, probably because of the decomposition of the Nabam in dilute solution.

The Zineb B treatment was about equal to the Nabam A treatment, but the Zineb A treatment and Maneb treatment were no better than tap water.

In the two experiments, there was a remarkable similarity in the numbers of larvae which emerged from the cysts in beet-root diffusate. The total number of larvae recovered (34,150 in the first and 34,200 in the second experiment) probably represents nearly the full potential number which could be hatched from these cysts. In similar experiments, Golden (6) reported weekly emergence of 7,520, 4,056, 1,292, 559, 171, and 80 larvae during the first to the sixth week, respectively. This is, 13,427 larvae of a total of 13,678, or more than 98%, hatched in the first four weeks, and less than 2% in the last two weeks.

The results can be summarized by stating that Nabam solutions containing 1,000 ppm increased hatching as compared with the tap water controls, but only about 60% as much as sugar beet-root diffusate. Addition of 1,197 ppm of manganese sulphate or 1,316 ppm of zinc sulphate reduced the action to about the same as tap water, whereas 658 ppm of zinc sulphate had no effect.

The increase in hatching in Nabam solutions is about the same as reported for ascorbic acid by Emanuelson (3), who considered the effect "not sufficiently pronounced to make it a suitable stimulus."

The reason for the increased hatching in Nabam solutions is not clear. It is highly unlikely that Nabam solutions contain the same "hatching factor" which occurs in beet-root diffusate or in ascorbic acid.

Dropkin et al. (2) have shown that hatching of nematode eggs can be delayed indefinitely by keeping them in solutions of high osmotic concentrations and that hatching proceeds when the osmotic stress is removed. These findings suggest that Nabam solutions and beet-root diffusate change the osmotic concentration in the egg in some way, perhaps by altering the permeability of the egg or larval membranes.

It can be concluded that while Nabam increases hatching, it is not highly efficient for the purpose in water solution. Its effect on *Heterodera* cysts in soil can only be determined by additional experiments, and its value for use in the field would depend on many factors including economic ones.

Summary

Laboratory tests were conducted to determine the effect of solutions of Nabam, Maneb, and Zineb on hatching of larvae from cysts of *Heterodera schachtii* as compared with those of tap water and beet-root diffusate during a 6-week period. In one test hatching in a solution containing 500 ppm of Nabam

was 42% of that in beet-root diffusate, 1,000 ppm 62%, 2,000 ppm 0.3% and in tap water 9%. In a second test, results were similar to these for 1,000 ppm of fresh Nabam solution and for 1,000 ppm of Nabam plus 658 parts zinc sulphate (Zineb B), but not different from the tap water controls for Maneb, 1,000 ppm of Nabam plus 1,316 ppm of zinc sulphate (Zineb A), or Nabam solution prepared six weeks before the beginning of the experiment.

Literature Cited

(1) CHRISTIE, J. R. 1959. Plant nematodes. Their bionomics and control. Agr. Exp. Sta. Univ. of Florida, Gainesville. p. 91.

(2) DROPKIN, V. H., MARTIN, G. C. and JOHNSON, R. W. 1958. The effect of osmotic concentrations on hatching of some plant-parasitic nematodes. Nematologica. 3: 115-126.

(3) EMANUELSON, H. 1954. An investigation of the sugar-beet eelworm, *Heterodera schachtii* (Schmidt). Socker Handlingar II. 8: 101-108.

(4) FRANKLIN, MARY T. 1951. The cyst-forming species of *Heterodera*. Commonwealth Agr. Bur. pp. 27-28.

(5) GOLDEN, A. M. 1958. Influence of leaf diffusate of sugar beet on emergence of larvae from cysts of the sugar-beet nematode (*Heterodera schachtii*). U. S. Dept. of Agr. Plant Dis. Reptr. 42: 188-193.

(6) GOLDEN, A. M. 1958B. Interrelationships of certain *Beta* species and *Heterodera schachtii*, the sugar-beet nematode. Plant Dis. Reptr. 42: 1157-1162.

(7) LUDWIG, R. A. and THORNE, G. D. 1953. Studies of the breakdown of disodium ethylene bisdithiocarbamate (nabam). U. S. Dept. of Agr. Plant Dis. Reptr. 37: 127-129.

(8) MILLER, P. M. and STODDARD, E. M. 1958. Increasing the hatching of eggs of cyst and root-knot nematodes with nabam. Science 128: 1429-1430.

(9) RASKI, D. J. and LEAR, B. 1958. Control of sugar-beet nematode. Calif. Agr. 12: 5-8 and 12.

Effects of Sugar Beet Nitrogen on Juice Purification

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A primary purpose of the sugar beet processing laboratory at the Western Regional Research Laboratory is the study of the effects of beet composition on processing characteristics (4)[2]. One of the most important changes in beet composition in recent years has been the increase in nitrogen content, presumably caused by increased use of inorganic fertilizers (3, 7, 9).

To test the effects of this increase on processing, ten samples of at least 100 beets each at six widely spaced sugar-to-total-nitrogen ratios were tested. All samples were grown under controlled conditions, eight samples in an experimental field at Woodland, California. The Woodland beets were grown by Drs. Lucile Hac and A. C. Walker of International Minerals and Chemical Company (IMC); the Berkeley beets by Dr. Albert Ulrich of the Division of Soils and Plant Nutrition, University of California. The eight Woodland samples included two varieties, US 75 and IMC 52, each grown with 100 lb or 300 lb nitrogen fertilization per acre. Variety IMC 52 was one developed by IMC for high glutamate values. The Berkeley beets of US 75 variety received ample water and minerals during growth, the main difference between the contrasted samples being the high and low quantities of nitrogen supplied.

Table 1 shows the effect of variety and nitrogen fertilization on yield, sugar, nitrogen, and glutamic acid. The field beets were harvested from two locations in the field with an interval of one week between harvest dates. The effects of harvest time and field location are thus inextricably confounded. It is interesting to note that the second harvest beets are uniformly higher in yield and sugar, and lower in nitrogen, than the first harvest beets, but that the effects of variety and fertilization are almost the same for the two harvest dates. Increased nitrogen fertilization results in higher yields of beets and sugar, with a slightly lower concentration of sugar. There is a large increase in total nitrogen,

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[2] Numbers in parentheses refer to literature cited.

Table 1.—Field effects of variety and nitrogen fertilization.

Fertilization		100# N/acre		300# N/acre		Low[1]	High[2]
Variety		US 75	IMC 52	US 75	IMC 52	US 75	US 75
Yield, T/A	A[b]	24.89	25.65	32.11	33.91	—	—
	B[b]	29.88	31.55	34.44	38.08		
Sugar, %	A	11.62	11.83	10.66	10.76	16.6	12.4
	B	11.95	11.92	11.72	11.69	—	—
Sugar, T/A	A	2.90	3.04	3.42	3.64		
	B	3.58	3.76	4.03	4.45		
Nitrogen, %	A	0.14	0.16	0.20	0.23	0.27	0.33
	B	0.13	0.15	0.18	0.23		
Nitrogen ×100 Sugar	A	1.2	1.35	1.9	2.1	1.6	2.7
	B	1.1	1.25	1.5	2.0		
Glutamic Acid, %	A	0.11	0.18	0.21	0.37	—	—
	B	0.10	0.19	0.24	0.43		

and an even larger increase in total glutamic acid with the higher fertilization level.

The beet samples were held in a moist room at 1° C for about seven days before slicing. Each entire sample was cut into standard cossettes, 100 g of which were 21 meters long. These were held about 3 hours at 1° C until diffused in the 12 kg per hour, Bruniche-Olsen continuous, countercurrent, single-scroll, laboratory diffuser (4). In each case the diffusion temperature was 70° C and the retention time 40 minutes. The efficiency of diffusion remained constant at 10.0 ± 0.4 theoretical contacts. About 1.8% of the sugar was lost in pulp which averaged about 0.2% sugar. No progressive change in pulp toughness by tenderometer was observed after 120 minutes equilibration. This suggests that diffuser retention time did not continuously increase and that holding the cossettes in the cold produced no important physical change.

The hot diffusion juices were carbonated immediately in the model Dorr carbonator (4, 5). All juices were carbonated at 80° C with 2.0% CaO based on beet weight using 7:1 recirculation to feed ratio of flows and 20 min retention time. Carbon dioxide was added as required to reach the following three test points: 0.08% CaO alkalinity; pH 10.15; and that which pro-

[1] Grown in pots at two levels of nitrogen, other nutrients in adequate amounts.
[2] A, first harvest; B, second harvest.

duced a settling rate of 18 lb mud per hour per square foot. When possible, samples of equilibrium effluent slurry at each of the three test points were collected for each run. Sedimentation and filtration rates were observed and thin juice was prepared from each sample. Second carbonation for this purpose consisted of adding carbon dioxide for 3 min to boiling first-carbonation filtrate followed by 5 min additional boiling. Analyses on the filtered juice included purity, lime salts, color, and total nitrogen. Specific nitrogen analyses were made for amide (10), amino (6) and ammonia (1).

Results of the purity determinations are shown in Figure 1 for diffusion juice and thin juice. Purity of diffusion juice and thin juice has an inverse relation to the beet nitrogen content for the field beets, but the pot grown beets yield juices with much higher purities than would be expected from the nitrogen level. The curves are least squares lines, r = —.97, for each juice from the field beets. There is no apparent difference in purity-nitrogen relationship between the varieties or harvest dates, but the pot beets are quantitatively different, although consistent in that the higher nitrogen results in lower purity. The small differences in purity for thin juices from a single sample of beets show no correlation with pH or alkalinity.

The extent of nitrogen removal during diffusion and carbonation, expressed as percent of original nitrogen, is reported in

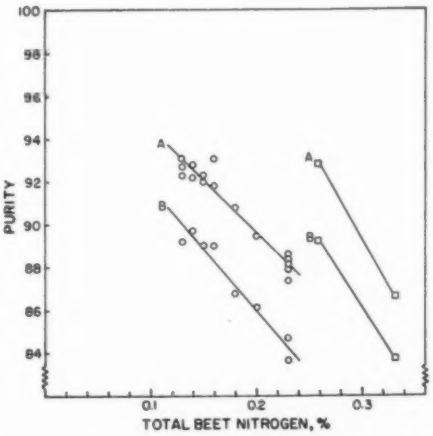

Figure 1.—Relation of total beet nitrogen to the purity of diffusion juice and thin juice produced in the processing laboratory. (A — thin juice, B — diffusion juice, O — field grown, □ — pot grown.)

Figure 2. Here there is a marked effect of harvest date-location, but two facts are apparent within harvest dates. The nitrogen removal by carbonation (circles) is a constant ratio of the original beet nitrogen, while the total amount removed by diffusion and carbonation (squares) is a smaller fraction at high nitrogen levels than at low nitrogen levels. The actual amount of nitrogen remaining in the pulp is almost constant for a particular harvest date. The nitrogen elimination during diffusion was 0.08, 0.06, and 0.09% on beet for the first harvest, second harvest, and pot grown beets, respectively. This can be explained by assuming a relatively constant level of protein in the beet, which means that an increase of nitrogen in the beet is reflected mainly in an increase in soluble nitrogen compounds. This tends to accentuate the differences in high and low nitrogen beets. Thus, in comparing the highest with the lowest nitrogen beets from the field plots, the nitrogen:sugar ratios are higher by 80% in the beet, 150% in the diffusion juice, and 180% in the thin juice. The total nonsugars do not increase in the same proportions, however, for the nonsugar:sugar ratios in diffusion juice and in thin juice are only 60% higher for the highest nitrogen beets than for the lowest nitrogen beets.

The soluble nitrogen in beets is largely from amino acids and betaine. As may be seen from Table 1, total glutamic acid increased faster than total nitrogen, under the influence of either fertilization or variety differences. Further evidence in this direction is shown in Figures 3 and 4, where the Van Slyke amino nitrogen as a percent of total nitrogen in diffusion juice or thin juice is plotted against original beet nitrogen. The regression line in Figure 3, with $r = 0.84$ ($p. = .01$), is for the field

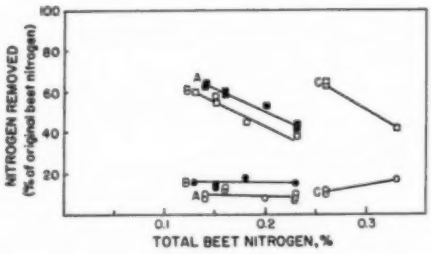

Figure 2.—Fraction of original nitrogen removed during diffusion and carbonation at various beet nitrogen levels (A — first harvest-location, B — second harvest-location, C — pot grown, ○ — removal during carbonation, □ — total removal during diffusion and carbonation.)

beets only. The dashed lines connect points representing a single variety and single harvest date. It is apparent that both variety and harvest date are related to the ratio of amino to total nitrogen, but the relation of beet nitrogen level is consistent in showing an increased proportion of amino nitrogen with increased total

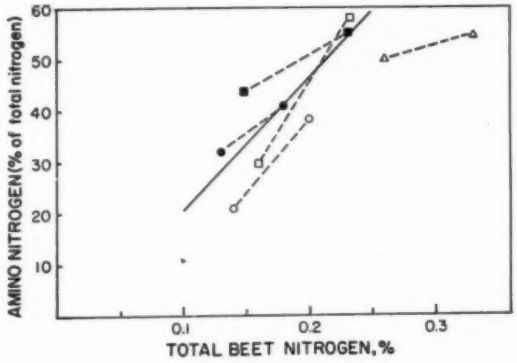

Figure 3.—Fraction of total nitrogen present as amino nitrogen in diffusion juice, related to beet nitrogen level, variety, and harvest date-location. (○ — US 75 first harvest-location, □ — IMC 52 first harvest-location, ● — US 75 second harvest-location, ■ — IMC 52 second harvest-location, Δ — US 75 pot grown.)

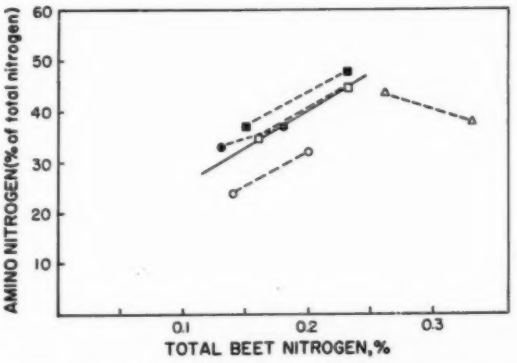

Figure 4.—Fraction of total nitrogen present as amino nitrogen in thin juice, related to beet nitrogen level, variety and harvest date-location (○ — US 75 first harvest-location, □ — IMC 52 first harvest-location, ● — US 75 second harvest-location, ■ — IMC 52 second harvest-location, Δ US 75 pot grown.)

nitrogen for each set of dates and variety. Amide and ammonia nitrogen vary with beet nitrogen in the same way as amino nitrogen. The quantity of each is about one fifth of the amino nitrogen level.

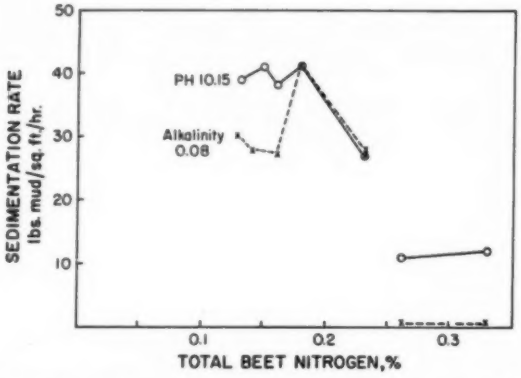

Figure 5.—First carbonation mud sedimentation rate at alkalinity 0.08 or pH 10.15 as a function of beet nitrogen level.

A similar picture is shown in Figure 4 for the thin juice, but the correlation coefficient is slightly lower, r= 0.77 (p = .05). In the case of thin juice, however, there is an analytical complication due to the formation of pyrrolidone carboxylic acid (PCA) from glutamine. PCA nitrogen does not contribute to the amino nitrogen value. Thus the juices with higher levels of glutamine will have a greater apparent loss of amino nitrogen during carbonation. The conversion of glutamine to PCA is not complete during carbonation, and does not necessarily occur to the same extent in different runs. Total and amino nitrogen concentrations were essentially unaffected by carbonation conditions.

The general conclusion is that as beet nitrogen increases, a larger proportion of the soluble nitrogen is amino nitrogen. Analyses for betaine were not made, but the inference is that betaine did not increase as fast as amino acid content, since betaine accounts for most of the soluble non-amino nitrogen.

Reducing sugar (2) in the diffusion juices from most samples was less than 0.1%. Carbonation removed 50% of the reducing sugar at 0.08 alkalinity, 70% at 0.10, and 80% at 0.12.

First carbonation was carried out at two standard points, one at alkalinity 0.08 (0.08% CaO) and the other at pH 10.15 (measured at 78° C with a glass electrode). As indicated earlier,

the different conditions of alkalinity or pH did not materially affect the thin juice purity or total nitrogen content. As would be expected, however, sedimentation rates and lime salts content are sensitive to carbonation conditions. Figure 5 shows the sedimentation rates at the two carbonation conditions as a function of beet nitrogen content. For levels of beet nitrogen below 0.18%, the alkalinity of 0.08 is too high for best sedimentation, and for the pot grown beets at beet nitrogen 0.26 and 0.33%, 0.08 alkalinity was too low for clarification to occur. For the pot grown beets it was necessary to raise the alkalinity to 0.11 to obtain a clear supernatant. This point corresponded to pH 10.00 at 78° C. Thus it appears that the buffering effect of the amino acids affects the alkalinity value at which first carbonation can be carried out successfully.

A further tentative conclusion which may be reached from the rather limited data gathered here is that above a beet nitrogen level of about 0.2%, there is an appreciable drop in the maximum sedimentation rate that can be reached. The points shown in Figure 5 do not represent all of the first carbonation conditions tried on these beets. Attempts were also made to determine the maximum pH and alkalinity at which first carbonation could be carried out and still give a sedimentation rate of 18 lb of mud per square foot per hour. This rate was calculated to be sufficient for average factory operation. This point was found for only three of the ten beet samples because of the limited sample size. The following observations were made from eight equilibrium points not included in Figure 5. All samples were consistent in showing a drop in sedimentation rate as the pH or alkalinity was raised above the point needed for clarification, but the amount of the drop in rate was related to the nitrogen content of the beets. Low nitrogen beets yielded juices much more sensitive to alkalinity changes than did the high nitrogen beets. For the low nitrogen US 75 beets the sedimentation rate dropped from 39 to 11 when the alkalinity rose from .06 to .12, while the rate for the high nitrogen IMC 52 juice dropped from 28 to 22 when the alkalinity was raised from .08 to .132. No conditions were found for any juice where the sedimentation rate was higher than it was at pH 10.15.

No simple trends of filterability or thin juice color with beet nitrogen were found. In the case of filterability, the coefficient of variation of 20% probably concealed any small true effects present (5). Color measurement had a coefficient of variation of 8% for equivalent treatments of one juice.

Lime salts content of thin juice is shown in Figure 6. Here it is apparent that the high-nitrogen field beets give much

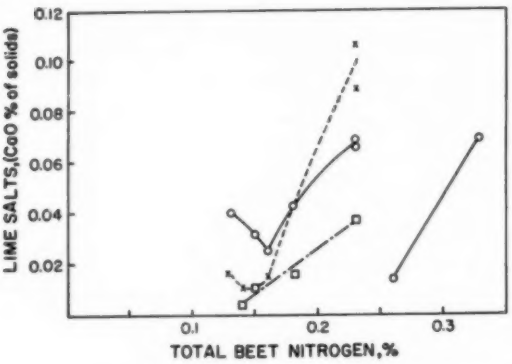

Figure 6.—Lime salts in filtered second carbonation juice from three first carbonation conditions as a function of beet nitrogen level. (X — alkalinity 0.08, O — pH 10.15, □ — sedimentation rate 15.22 lb mud/sq ft/hr).

higher lime salts than the low-nitrogen beets. No points are shown for the pot grown beets at alkalinity .08 because no thin juice was produced where the first carbonation juice could not be clarified by settling. The very low lime salts content of the lower nitrogen pot grown beets does not fit in with the picture of high lime salts with high-nitrogen content, but again illustrates the difference in behavios of field beets and pot beets. In general a reduction of lime salts was found at higher alkalinity or pH levels. When the alkalinity was raised until the sedimentation rate had dropped to 15-22 lb/sq ft/hr, the lime salts were in direct relation to the beet nitrogen.

Before assessing the economic effect of various nitrogen levels in beets, more data would be required, particularly on the amount of molasses produced. The findings reported here show clearly that high levels of nitrogen fertilization result in juices of low purity and high nitrogen content. In a study of molasses composition and purity (8) a high negative correlation was found between nitrogen content and purity for straight house molasses, indicating that nitrogen compounds may be less melassigenic than the average of other impurities. This is not to say that high nitrogen juices would yield less molasses than low nitrogen juices, but that the increase in molasses due to low purity thin juices in the present study could not be accurately predicted from the purity figures alone.

Summary

The nitrogen level in beets is related to the nitrogen fertilization and to the variety of the beets. Characteristics affected by nitrogen level of the beet are purity, soluble nitrogen, lime salts, and sedimentation rate. All of these factors are adversely affected by increased nitrogen concentration. The level of nitrogen in the juice also affects the alkalinity, but not the pH, required for optimum first carbonation. Additional data would be needed to predict the amount of additional molasses produced from high nitrogen beets.

The authors wish to thank Taysir Jaouni and Robert Patterson for their assistance in the processing laboratory, and Marion Long, Henry Wright and Earl Potter for the analyses for nitrogen, total solids, and reducing sugars. Thanks are also due to Dr. Albert Ulrich for beet samples, and to Dr. Lucile Hac and Dr. A. C. Walker for beet samples and for data on yield and glutamic acid content of the field beets.

Literature Cited

(1) Association of Official Agricultural Chemists. 1955. Official Methods of Analysis, 8th Ed., p. 371.

(2) *Ibid.*, p. 119.

(3) Haddock, J. L., Smith, P. B., Downie, A. R., Alexander, J. T., Easton, B. E., and Jensen, V. 1959. The influence of cultural practices on the quality of sugar beets. J. Am. Soc. Sugar Beet Technol. 10: 290-301.

(4) Morgan, A. I., Barta, E. J., and Kohler, G. O. 1959. Development of a sugar beet processing laboratory. J. Am. Soc. Sugar Beet Technol. 10: 563-570.

(5) Morgan, A. I., Goodban, A. E., Teranishi, R., Knowles, R. E., and McCready, R. M. 1958. Effects of some variables on first carbonation. J. Am. Soc. Sugar Beet Technol. 10: 396-402.

(6) Peters, J. P., and Van Slyke, D. D. 1946. Quantitative Clinical Chemistry, 2nd Ed., Williams and Wilkins Co., Balt. Vol. 1, Pt. 1.

(7) Rounds, H. G., Rush, G. E., Oldemeyer, D. L., Parrish, C. P., and Rawlings, F. N. 1958. A study and economic appraisal of the effect of nitrogen fertilization and selected varieties on the production and processing of sugar beets. J. Am. Soc. Sugar Beet Technol. 10: 97-116.

(8) Stark, J. B., McCready, R. M., and Sandomire, M. M. 1960. Non-sugars in molasses and their influence on purity. Presented at meeting of Am. Soc. Sugar Beet Technol., Salt Lake City, Utah, Feb. 1960.

(9) Stout, Myron. 1961. A new look at some nitrogen relationships affecting the quality of sugar beets. J. Am. Soc. Sugar Beet Technol. 11: 388-398.

(10) Winton, A. L. 1945. Analysis of Food, John Wiley & Sons, New York, p. 549.

Testing of Inbred Lines of Sugar Beets for Resistance to Aphanomyces, Rhizoctonia and Fusarium Root Rots[1]

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A large number of open pollinated and inbred lines of sugar beets have been tested for their resistance to Aphanomyces, Rhizoctonia and Fusarium root rots since 1950 (4, 5, 6)[3].

In earlier work in this laboratory, many open pollinated lines of beets were tested for their resistance to these diseases. Later on, studies were conducted mainly with inbred lines of beets since they represent a population of greater genetic uniformity. Seeds of such open pollinated lines of beets were received, mainly from various commercial sugar beet companies. Seeds of inbred lines of beets were supplied through the Beet Sugar Development Foundation.

In all tests a mixture of inoculum was used for each of the following pathogenic organisms: *Aphanomyces cochlioides* Drechs.; *Pellicularia filamentosa* (Pat.) Rogers, (*Rhizoctonia solani);* and *Fusarium oxysporum* f. *betae* (D. Stewart) Snyder and Hansen. Pure cultures of these organisms were isolated from infected sugar beets. The virulence of these cultures was determined regularly in separate tests.

The inocula for these tests usually were prepared by growing the fungi in petri dishes containing potato dextrose or oatmeal agar. After a sufficient amount of growth developed, it was scraped off the agar, homogenized in a blender and added to steam-sterilized soil in pots immediately after beet seeds were planted. Usually one petri dish of inoculum was used per seven- or eight-inch pot of soil.

In most of the testing, the following technique was used: All beet lines were submitted first to a screening test for three root rots (6). Those lines which showed a good degree of resistance to any of these diseases in the screening test were retained for further testing. After the first cycle of selection, five apparently resistant beets from each line which showed resistance were either selfed or sib-mated depending on whether they were self fertile or self sterile. The seeds produced from these beets were tested again for resistance to the same disease for which the line

[1] Contribution from Montana State College, Agricultural Experiment Station, Bozeman, Montana. Paper No. 482.

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[3] Numbers in parentheses refer to literature cited.

was selected and only those lines of beets were retained which showed an outstanding degree of resistance.

After the second cycle, resistant beets from self-fertile lines were selfed while resistant beets from self-sterile lines were grouped according to which disease they were resistant to and cross pollinated. An attempt was made to produce a large amount of seed from these beets in order to perform large-scale tests either in the field or in the greenhouse. Prior to large-scale testing, all lines of beets obtained through cross pollination were again tested in the greenhouse and all susceptible entries were discarded.

In the first two cycles of testing, resistance of beets to each respective root rot disease was tested in the seedling stage and beets were harvested usually about six weeks after emergence. During the last test, when they were tested on a large scale, they were allowed to grow for several months in order to test for resistance at an older plant stage. Readings of healthy and diseased beets were always taken during the growing season and at harvest. Table 1 gives coding identification and source for various sugar beet entries reported in this paper.

Table 1.—Coding system and sources for the various sugar beet entries.

Code	Source
US	USDA, Beltsville, Maryland
SP	USDA, Beltsville, Maryland
SL	U.S. Sugar Plant Field Laboratory, Salt Lake City, Utah
SLC	U.S. Sugar Plant Field Laboratory, Salt Lake City, Utah
D	USDA, Fort Collins, Colorado
FC	USDA, Fort Collins, Colorado
GW	The Great Western Sugar Company, Denver, Colorado
ACS	American Crystal Sugar Company, Denver, Colorado
HSC	Holly Sugar Corporation, Colorado Springs, Colorado
TASCO	The Amalgamated Sugar Company, Ogden, Utah
AC	Michigan State University, East Lansing, Michigan

Aphanomyces Root Rot

None of the tested lines of sugar beets consistently showed a high degree of resistance to Aphanomyces root rot in the greenhouse tests. Greater differences in disease resistance between lines may have been detected at a lower concentration of inoculum. Even though there does not appear to be a specific resistance among these lines of beets to the disease, some beet lines were more resistant than others and beets may become infected and still recover sufficiently to give a satisfactory stand. There was

some variation between tests in disease ratings for the inbred lines and their progenies. These variations may have been due to some differences in the growing conditions and in the concentration of inoculum.

The following eight lines of beets showed a higher degree of resistance to Aphanomyces root rot than the others:

US acc - 1241	SP5270-0
D1952-359	GWI-42
D1952-441	GWI-44
SP5263-0	HSC5126-0

Under field conditions, beet lines were infected considerably less than in the greenhouse. This difference may have been due to lower inoculum concentrations in the field or to a difference in other environmental factors. It is quite possible that by exposing beets to a lower standardized inoculum in the greenhouse, similar to the method used by Schneider (7), one may be able to obtain a closer correlation between greenhouse and field tests. More work is required to clarify this situation.

In early work with Aphanomyces root rot it was shown that the seedling phase of this disease can be controlled fairly well in the field by improving the physical conditions of the soil and by providing sufficient and balanced fertilization (1, 2, 3). It is possible that this disease can be controlled satisfactorily by planting beet lines with a moderate degree of resistance to Aphanomyces root rot in soil well supplied with organic matter and fertilizers.

At the present time the supply of seed of the eight abovementioned beet lines, is being increased so that further tests can be conducted.

Rhizoctonia Root Rot

Most of the following 16 lines of beets showed only a moderate degree of resistance to Rhizoctonia root rot. The concentration of inoculum was undoubtedly an important factor in the performance of these tests. It appears that these lines of beets also do not have a specific resistance to this disease. Variation in disease ratings between tests was similar to that observed for Aphanomyces root rot:

AC9-406-0	SLC238	D1954-600	ACS-I_1
SL2-501-1-4	D1941-164	FC55-8055	ACS-I_3
SL028	D1950-351	SP5478-0	HSC5152-0
SLC207	D1950-540	GWI-5	TASCO 6-284

Fusarium Root Rot

A rather large number of beet lines showed a moderate to high degree of resistance to Fusarium root rot. When the original

seed or their progenies was tested, less variation in degree of resistance occurred for this disease than for Aphanomyces or Rhizoctonia root rots. It appears that resistance of these lines of beets to Fusarium root rot may be more specific in nature.

The following twenty-five lines of beets showed a fairly high degree of resistance to Fusarium root rot:

SL509	D1951-500	D1953-337	FC55-8022
SLC238	D1952-388	D1954-315	FC55-8034
D1950-444	D1952-393	D1954-593	AC32:57302
D1950-558	D1952-443	D1954-467	AC310:851909
D1951-319	D1952-444	FC55-8027	GWI-8-55L
D1951-356	D1953-328	FC55-8057	GWI-43
			ACS-14

Further tests will be conducted with those lines which showed the highest degree of resistance to each of the three root rots discussed.

It appears that the use of a standard concentration of inoculum is an important factor in the evaluation of the resistance of beet lines. Evidently the concentration of the inoculum used in these tests was considerably higher than in the disease exposure which beets encountered under ordinary field conditions. In further tests, standardization of inoculum will be made and especially in regard to testing beets for resistance to Aphanomyces root rot.

Screening of beet lines and selection within varieties has been most successful for Fusarium root rot, followed by Rhizoctonia and Aphanomyces. However, the development of a Rhizoctonia resistant varieties of beets would be most desirable since this disease causes the greatest loss in the Western sugar beet areas. Some inherent resistance apparently exists for Rhizoctonia and Aphanomyces root rots, but multigenic inheritance probably decreases the possibility of obtaining highly resistant varieties. Concentration of resistance genes by recurrent mass selection might result in development of beet varieties with higher degree of resistance to these root rots than the varieties presently grown.

Literature Cited

(1) AFANASIEV, M. M. and MORRIS, H. E. 1943. Diseases of sugar beets in crop rotations at Huntley Branch Station, Huntley, Montana, from 1936 to 1941. Mont. Agr. Exp. Sta, Tech. Bull. 419.

(2) AFANASIEV, M. M. 1948. The relation of six groups of fungi to seedling diseases of sugar beets in Montana. Phytopath. 38 (3) : 205-212.

(3) AFANASIEV, M. M. and MORRIS, H. E. 1949. Effect of fertilization on the recovery of transplanted sugar beet seedlings affected with *Aphanomyces cochlioides* Drechs. in the greenhouse. Phytopath. 39 (12) : 1001-1004.

(4) AFANASIEV, M. M. and MORRIS, H. E. 1952. Resistance and soil treatments for control of Rhizoctonia of sugar beets. Proc. Am. Soc. Sugar Beet Technol. 7: 562-567.

(5) AFANASIEV, M. M. and MORRIS, H. E. 1954. Testing sugar beet varieties for their resistance to Aphanomyces, Rhizoctonia, and Fusarium root rots. Proc. Am. Soc. Sugar Beet Technol. 8 (2) : 90-93.

(6) AFANASIEV, M. M. 1956. Resistance of inbred varieties of sugar beets to Aphanomyces, Rhizoctonia, and Fusarium root rot. J. Am. Soc. Sugar Beet Technol. 9 (2) : 178-179.

(7) SCHNEIDER, C. L. 1954. Methods of inoculating sugar beets with *Aphanomyces cochlioides* Drechs. Proc. Am. Soc. Sugar Beet Technol. 8 (1) : 247-251.